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McClellan AFB Soil Vapor Extraction (SVE) Off-Gas Technology Selection Characterization, Literature Review, and Technology Selection

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14. Abstract
McClellan AFB has four soil vapor extraction (SVE) systems operating to remediate soil contaminated with chlorinated organic compounds. Treatment of the exhaust gases from these systems is expensive and could be enhanced by using a bioreactor treatment system. Following a chemical analysis of the contaminants in the SVE off-gases, a literature search was conducted to identify the scientific basis of several treatment options. A reference list of 175 sources is provided. The various types of available bioreactors and the treatment processes used are discussed along with the rationale for the selection of a biotrickling filter system to be tested in the laboratory prior to installation at McClellan AFB.

15. Subject Terms bioreactor, biotrickling filter bioreactor, BTEX, chlorinated aliphatic compound, chlorinated aromatic compound, soil vapor extraction off-gas, tetrachloroethene (PCE), trichlorothylene (TCE), site remediation

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SUMMARY

The United States Air Force is responsible for the cleanup of environmental contamination that exists on Air Force installations/property. In order to remediate subsurface contamination, both unsaturated and saturated, numerous in situ remediation techniques have been developed. One of those techniques is Soil Vapor Extraction (SVE). McClellan AFB, California, has installed four SVE systems to remediate vadose zone contamination. The resultant off-gas streams are currently remediated by catalytic oxidative processes. Catalytic oxidative off-gas treatment systems are expensive to operate. A more effective, efficient, and inexpensive off-gas treatment is desired. Bioreactors offer a promising solution, which may be applicable at McClellan AFB.

Gas samples from the McClellan AFB SVE systems that were in use in Operating Unit D (OU D) were collected and analyzed to determine the types of contaminants and their concentrations. The data generated from this analysis were used to select the appropriate bioreactor technology for use at McClellan AFB. This technology would be tested in the laboratory to verify its effectiveness before a field test plan would be written and a field demonstration started. The off-gas consisted of chlorinated aliphatic, chlorinated aromatic, and nonchlorinated organic compounds. The average SVE flow rate for all wells in OU D was 1,045 scfm. The compounds that were tested in the laboratory phase included an aerobic-cometabolically degraded chloroethene (TCE), an anaerobically-degraded chloroethene (PCE), an aerobic-cometabolically degraded chloroethane (1,1,1-TCA), an aerobically degraded dichlorobenzene (1,2-DCB), an aerobically degraded BTEX compound (toluene), and acetone, which is aerobically degraded.

A large portion of this report reviews the microbiology of off-gas contaminant degradation, including chlorinated and nonchlorinated compounds. Particular attention was paid to aerobic cometabolic degradation, because that is the biological mechanism that will be used to degrade most of the chlorinated organic compounds in the off-gas stream. The report also contains a discussion of the types of bioreactors that are available, the process parameters that can be used within the bioreactor, and the rationale for the selection of the biotrickling filter as the most suitable reactor for further laboratory testing before installation as the McClellan AFB SVE off-gas treatment system. The reference section of the report contains 175 entries which provide detailed information on the microbiology of off-gas contaminant degradation.

LIST OF ACRONYMS

AFB	Air Force Base
BTEX	benzene, toluene, ethylbenzene, and xylenes
CAA	Clean Air Act Amendments
CAC	chlorinated aliphatic compound
Cal/EPA	California Environmental Protection Agency
CF	chloroform
COC	chlorinated organic compound
CT	carbon tetrachloride
DCA	dichloroethane
DCB	dichlorobenzene
DCE	dichloroethene (dichloroethylene)
DCM	dichloromethane
DCP	dichloropropylene
DO	dissolved oxygen
DoD	U.S. Department of Defense
DRE	destruction removal efficiency
EPIC	Environmental Process Improvement Center
FBR	fluidized-bed reactor
FTO	flameless thermal oxidation
GC	gas chromatograph
HCl	hydrochloric acid
HF	hydrofluoric acid
IPB	isopropylbenzene
MIBK	methyl isobutyl ketone
MEK	methyl ethyl ketone
MMO	methane monooxygenase
MW	molecular weight
NaCl	sodium chloride
NADH	nicotinamide adenine dinucleotide
NCOC	nonchlorinated organic compound
NETTS	National Environmental Technology Test Site

NJDEP	New Jersey Department of Environmental Protection
NO_x	Nitrous oxides
NPD	nonthermal plasma destruction
OU	operating unit
PAC	powdered activited carbon
PAH	polycyclic aromatic hydrocarbon
PCE	tetrachloroethene (tetrachloroethylene perchloroethylene)
PD	photolytic destruction
pMMO	particulate MMO
QA/QC	quality assurance/quality control
RA	regenerable adsorption
Rs	cosubstrate/contaminant feeding ratio
SD	standard deviation
SERDP	Strategic Environmental Research and Development Program
sMMO	soluble MMO
SO_x	Sulfur oxides
SVE	soil vapor extraction
TAC	toxic air contaminant
TCA	trichloroethene (trichloroethane)
TCE	trichloroethylene
TeCA	tetrachloroethane
TMB	trimethylbenzene
TNMOC	total nonmethane organic compound
TOC	total organic carbon
TOM	toluene *ortho*-monooxygenase
TOPD	titanium oxide photocatalytic destruction
TPH	total petroleum hydrocarbons
U.S. EPA	U.S. Environmental Protection Agency
UV	ultraviolet
VC	vinyl chloride (chloroethylene)
VOC	volatile organic compound
VSS	volatile suspended solids

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SECTION I

INTRODUCTION

McClellan Air Force Base (AFB) has been designated as the Chlorinated Hydrocarbons Remedial Demonstration Site as part of the National Environmental Technology Test Site (NETTS) program.

The current process for gaining commercial acceptance of cost-effective, innovative technologies for the cleanup of federal installations is laborious and costly. The NETTS program was established in 1993 to facilitate the transition of environmental remediation technologies to full-scale use by overcoming the barriers that presently inhibit commercialization of such technologies. NETTS is a joint Department of Defense (DoD) and U.S. Environmental Protection Agency (U.S. EPA) program funded by the Strategic Environmental Research and Development Program (SERDP).

A. OBJECTIVES

The overall objectives of this study are to identify biologically mediated methods for the treatment of VOCs in air waste streams resulting from in situ environmental remediation technologies at McClellan AFB; demonstrate the effectiveness of selected technologies in the laboratory; and perform field demonstrations of the selected technologies.

B. SCOPE

The work is divided into two phases: Phase I includes the review and characterization of the existing soil vapor extraction (SVE) off-gas based on existing data, review of current literature and available technologies, selection of one technology for further study, and preparation of a technology implementation plan for laboratory and field demonstrations of the selected technology. Phase II is divided into two parts, Phase IIA and Phase IIB, and will involve the execution of the technology implementation plan prepared in Phase I. Phase IIA will include a laboratory demonstration of the selected technology, and Phase IIB will include a field demonstration of the same technology. The objectives of this Phase I deliverable report are to present the off-gas characterization results, to review the pertinent literature for biological treatment of the off-gas organic contaminants, and to select a process technology to be demonstrated in the laboratory. The target destruction removal efficiency (DRE) for the organic contaminants in the McClellan AFB SVE off-gas is 95% of the total amount.

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SECTION II

SVE OFF-GAS CHARACTERIZATION

Raw data were obtained from the Air Force for the purpose of characterizing the individual organic contaminant concentrations in the SVE off-gas stream. The samples were collected between August 1995 and March 1996, from a sampling port in a manifold section that conducted off-gas from expansion wells at Sites A and J, and 8 newly installed monitoring wells in Sites 2, 4, and 5 in Operating Unit D (OU D). The eight wells were expected to produce relatively high off-gas contaminant concentrations and to represent a "worst-case scenario" for the base. Off-gas organic contaminant characteristics are summarized in Section III A with respect to individual organic contaminant concentrations and mass flow rates. The bioreactor technology selection is based on this data set. Because the system was operating under "worst-case" conditions, the data are expected to result in a conservative selection and design of the bioreactor for the removal and destruction of contaminants in the SVE off-gas.

The off-gas consists of chlorinated aliphatic, chlorinated aromatic, and nonchlorinated organic compounds. TO-14 or EPA 8010/8020 analytical methods were used to quantify the off-gas contaminants; all three methods test for a wide range of organic compounds. Contaminant concentrations measured in the off-gas at OU D, between August 1995 and March 1996, are shown in Table 1; only those organic contaminants that were measured in the off-gas above the method detection limits are shown. The contaminants in the off-gas include chloroethenes, chloroethanes, chlorobenzenes, chloromethanes, trimethylbenzenes, Freon™ 113, benzene, toluene, ethylbenzenes, and xylenes (BTEX) compounds, acetone, and ketones.

Table 1 shows the minimum, maximum, and average concentrations measured in 9 to 29 samples. The number of samples for each contaminant depended on the method employed; method TO-14 measured for a wider range of contaminants than EPA-8010/8020. The number of sample hits for each compound also is shown. Some samples were consistently detected; for example, trichlorothylene (TCE) was detected in 29 out of 29 samples. Other samples were detected less frequently; for example, chloromethane was detected in 1 out of 9 samples, and vinyl chloride (VC) was detected in 1 out of 28 samples. The minimum, maximum, and average off-gas concentrations are based on the actual sample hits rather than on the total number of samples. The standard deviation (SD) represents the deviation of the same set of samples.

The average total SVE flow rate for all the samples was 1,045 scfm and was used to determine the mass flow rate in pounds of contaminant per hour (lb/hr). Aqueous-phase concentrations (mg/L) were determined using the average gas-phase concentrations and their respective Henry's law constants (atm-m^3/mole), assuming saturated conditions. Henry's law constants are shown in Table 2, along with other physical constants including molecular weight (MW), boiling point (^oC), solubility (g/L), and octanol-water partition coefficient (K_{ow}).

Chloroethenes measured in the off-gas included tetrachloroethene (PCE), TCE, 1,1-dichloroethene (1,1-DCE), *cis*-1,2-dichloroethene (*c*-DCE), and VC. TCE had the highest maximum (111 ppmv) and average concentrations (74 ppmv) of all the chloroethenes in the off-gas, followed by PCE (97 and 63 ppmv, respectively). The other chloroethene concentrations were below 10 ppmv.

Chloroethanes detected in the off-gas included 1,1,1-trichloroethane (1,1,1-TCA), 1,1,2-TCA, and 1,1-dichloroethane (1,1-DCA). The 1,1,1-TCA appeared at the highest maximum (241 ppmv) average concentrations (152 ppmv) of all the chlorinated aliphatic compound (CACs) detected in the off-gas. The other chloroethane concentrations were below 10 ppmv.

Of the dichlorobenzenes (DCBs) detected in the off-gas (1,2-, 1,3-, and 1,4-DCB), 1,2-DCB had the highest maximum (121 ppmv) and average concentrations (35.5 ppmv). The average concentrations of the other DCBs were below 10 ppmv. Chlorobenzene also was detected at relatively low concentrations (1.0 ppmv average).

Chloromethane and methylene chloride both were detected below 10 ppmv, and the only fluorinated compound, Freon™ 113, was detected with an average concentration of 1.3 ppmv.

Of the nonchlorinated compounds, BTEX compounds totaled 66.5 ppmv, acetone concentrations averaged 63.8 ppmv, methyl ethyl ketone (MEK) averaged 7.3 ppmv, and methyl isobutyl ketone (MIBK) averaged 17.5 ppmv. Of the BTEX compounds, toluene had the highest average concentration of 45.5 ppmv.

The total sum of the average chlorinated organic compound (COC) concentrations was 374 ppmv, resulting in a total aqueous-phase concentration of approximately 10 mg/L. The total sum of the average nonchlorinated organic compound (NCOC) concentrations was 160 ppmv, resulting in a total aqueous-phase concentration of approximately 242 mg/L. The very high aqueous-phase NCOC concentration is due primarily to the high projected acetone concentration of 180 mg/L resulting from acetone's high solubility and low Henry's constant.

TABLE 1. OFF-GAS CONTAMINANT CHARACTERIZATION

Compound	Off-gas Concentration Min. (ppmv)	Max. (ppmv)	Avg. (ppmv)	n/total	SD	Mass Flowrate (lb/hr)	Conc. in H_2O (mg/L)
PCE	34.7	97.4	63.0	28/28	13.19	1.70	0.403
TCE	28.3	111.3	74.4	28/28	21.71	1.59	1.07
c-DCE	1.0	3.8	2.3	24/29	0.74	0.04	0.068
1.1-DCE	1.7	5.8	3.6	23/29	1.12	0.056	0.012
VC	0.2	0.2	0.2	1/28		0.002	0.001
1.1.1-TCA	53.9	241.4	152.9	29/29	46.64	3.321	4.15
1.1.2-TCA	0.8	0.8	0.8	1/9		0.018	0.092
1.1-DCA	1.6	5.2	3.6	28/29	0.89	0.057	0.065
1.2-DCB	1.2	120.6	35.5	27/29	34.57	0.849	2.82
1.3-DCB	1.4	5.2	2.8	5/9	1.54	0.068	0.235
1.4-DCB	1.6	21.5	6.6	8/9	6.26	0.157	0.651
Chlorobenzene	0.5	2.3	1.0	14/29	0.40	0.019	0.034
1.2.4-TMB	1.0	38.9	16.3	29/29	12.43	0.318	0.279
1.3.5-TMB	2.2	4.0	3.1	8/9	0.72	0.061	0.062
Methylene Chloride	1.9	7.8	5.4	28/28	1.73	0.074	0.225
Chloromethane	1.7	1.7	1.7	1/9		0.014	0.004
Freon™ 113	0.9	1.8	1.3	8/27	0.31	0.042	0.001
Benzene	0.1	5.4	2.8	2/29	3.77	0.035	0.040
Toluene	19.1	76.8	45.5	28/28	14.02	0.681	0.657
Ethylbenzene	1.2	8.3	3.6	27/28	1.81	0.062	0.046
Xylenes, Total	5.9	30.0	14.6	28/28	5.49	0.252	0.220
4-Ethyl Toluene	2.7	6.7	4.6	9/9	1.23	0.090	
Acetone	24.6	92.0	63.8	9/9	18.84	0.602	180
Methyl Ethyl Ketone	7.3	7.3	7.3	1/9		0.085	19.14
Naphthalene ($\mu g/m^3$)	1359	1678	1518	2/8	225	0.006	0.06
2-Methylnaphthalene ($\mu g/m^3$)	628	764	696	2/8	96	0.003	9.0×10^{-4}
Anthracene-d ($\mu g/m^3$)	28.91	34.41	31.68	2/8	3.85	1.2E-4	0.027
Pyrene-d ($\mu g/m^3$)	21.04	23.90	22.47	2/8	2.02	8.8E-05	NA
Terphenyl-d ($\mu g/m^3$)	2.20	3.06	2.63	2/8	0.61	1E-05	NA
Methyl Isobutyl Ketone	6.5	21.1	17.5	9/9	4.73	0.285	42.08
Chlorinated Sum	131.4	626.8	355.1			8.0	9.8
Nonchlorinated Sum	67.3 [(a)]	247.6 [(a)]	159.5 [(a)]			2.1	242.3

Average Flowrate = 1044.8 scfm during sampling.

(a) Nonchlorinated sum of off-gas concentrations do not include PAH compounds measured in $\mu g/m^3$.

TABLE 2. PHYSICAL CONSTANTS FOR CONTAMINANTS DETECTED IN THE OFF-GAS

Compound	MW	Boil. Pt (°C)	Solubility (g/L) (in H_20)	H (atm m^3 /mol) (@25C)	K_{ow}
PCE	165.83	121	0.15	2.59E-02	398
TCE	131.39	87	1.10	9.10E-03	194.98
c-DCE	96.94	60.3	3.50	3.37E-03	1.86
1,1-DCE	96.94	32	2.40	2.96E-02	134.9
VC	62.50	-13	2.80	1.09E-02	23.9
1,1,1-TCA	133.40	74	4.40	4.92E-03	309
1,1,2-TCA	133.40		4.50	1.17E-03	295
1,1-DCA	98.96	57.5	5.50	5.43E-03	61.7
1,2-DCB	147.00	180	0.15	1.85E-03	2398.8
1,3-DCB	147.00	173	0.11	1.78E-03	3981.1
1,4-DCB	147.00	174	0.08	1.48E-03	3311.3
Chlorobenzene	112.56	132	0.49	3.46E-03	691.80
Methylene Chloride	84.93	40	20.00	2.03E-03	20
Chloromethane	50.49	-24	NA	2.37E-02	8.13
Freon™ 113	187.40	NA	0.18	0.178	2000
1,2,4-TMB	120.19	170	NA	7.00E-03	NA
1,3,5-TMB	120.19	162 to 164	0.02	6.00E-03	NA
Benzene	78.11	80	1.77	5.40E-03	134.9
Toluene	92.04	111	0.53	6.37E-03	537
Ethylbenzene	106.17	136	0.16	8.39E-03	1412.5
Xylenes, Total	106.17	138 to 144	0.16-0.18	7.04E-03	318.3 to 584.9
4-Ethyl Toluene	120.20	162	NA	NA	NA
Acetone	58.00	56.2	imisible	2.06E-05	0.58
Naphthalene	128	218	NA	6.1×10^{-4}	NA
Methylnaphthalene	142	244.6	NA	0.18	NA
Acenapthalene	152	265	NA	12.8×10^{-5}	NA
Methyl Ethyl Ketone	72.12	79.7	2.68	2.74E-05	1.88
Methyl Isobutyl Ketone	100.16	117.5	NA	4.16E-05	5.25

Source: Multiple Sources

Table 3 shows the COCs and NCOCs that will be used for the laboratory demonstration phase. The compounds shown in Table 3 were selected for two primary reasons. First, we selected organic contaminants that would represent the different groups of contaminants detected in the off-gas stream (i.e., chloroethenes, chloroethanes, chlorobenzenes, BTEX compounds, and acetone). The second criterion was to select the compounds that appeared at the highest concentrations in the off-gas. The compounds that were selected comprise approximately 85% of the total average mass flow rate in Table 1. The compounds that will be tested in the laboratory phase include an aerobic-cometabolically degraded chloroethene (TCE), an anaerobically-degraded chloroethene (PCE), an aerobic-cometabolically degraded chloroethane (1,1,1-TCA), an aerobically degraded dichlorobenzene (1,2-DCB), an aerobically degraded BTEX compound (toluene), and acetone, which also is aerobically degraded. VC was not included because it represents a minor fraction of the CACs in the off-gas. PCE was selected because it requires anaerobic conditions to be degraded. TCE and TCA were selected as representative chloroethene and chloroethane constituents. DCB was selected as a representative chlorinated aromatic compound. Toluene was selected as a representative nonchlorinated aromatic compound, and because it could contribute to the cometabolic degradation of the CACs. Acetone was selected as a representative nonchlorinated aliphatic compound, and because of its high concentrations in the gas stream. The total number of compounds is limited to six due to analytical limitations and limited time and funds.

TABLE 3. CONTAMINANTS SELECTED FOR THE LABORATORY DEMONSTRATION ((a) assumes flow rate of 1 L/min (0.035 ft^3/min))

	Off-gas Conc.			Mass Flow	Conc. in
	Max.	Avg.		Rate[(a)]	H_2O
Compound	(ppmv)	(ppmv)	n/total	(10^{-4} lb/hr)	(mg/L)
PCE	97.4	63.0	28/28	2.73	0.403
TCE	111.3	74.4	28/28	3.23	1.07
1,1,1-TCA	241.4	152.9	29/29	6.63	4.15
1,2-DCB	120.6	35.5	27/29	1.54	2.82
Toluene	76.8	45.5	28/28	1.97	0.657
Acetone	92.0	63.8	9/9	2.77	180
Chlorinated Sum	570.70	325.80		14.74	8.44
Nonchlorinated Sum	168.8	109.3		4.74	180.7

Trimethylbenzenes and ketones are not represented in Table 3. Those compounds also are expected to degrade slowly. Freon™ 113 is another biologically recalcitrant compound that has been detected in the off-gas stream. Freon™ 113 concentrations in the off-gas averaged 1.3 ppmv, resulting in a projected aqueous-phase concentration of 0.001 mg/L.

Biological degradation of the halogenated compounds in the off-gas will result in the production of hydrochloric acid (HCl) and hydrofluoric acid (HF) compounds, which could produce acidic conditions in the wastestream. In the laboratory-scale demonstration, the estimated HCl released into the reactors will be 1.8 to 3.6 g Cl^-/day (0.05 to 0.10 mol/day), assuming (1) a gas flow rate of 0.05 to 0.1 scfm, (2) the target COC concentrations shown in Table 3, and (3) complete mineralization of all the organic chlorine. This high release of HCl and HF compounds will require careful pH control and/or high alkalinity concentrations to buffer the pH of the system. Otherwise, the HCl and HF will result in acidic conditions that will be toxic to the bacteria, as well as corrosive to the system.

SECTION III

LITERATURE REVIEW

This section reviews the microbiology of off-gas contaminant degradation, including chlorinated and nonchlorinated compounds. Particular attention is given to aerobic cometabolic degradation, because that is the biological mechanism that will be used to degrade most of the COCs in the off-gas stream.

A. DEGRADATION OF SVE OFF-GAS CONTAMINANTS: MICROBIOLOGY

This section focuses on the microbiology of contaminant degradation in the SVE off-gas. The results of the off-gas characterization show that the SVE off-gas consists of a very complex mixture of contaminants. It is important to understand the limitations, as well as the potential advantages, of the different biological processes for degrading the off-gas contaminants. The degradation characteristics of the off-gas contaminants are divided into the following groups: COCs degraded cometabolically, COCs and NCOCs degraded as primary growth substrates, and COCs degraded anaerobically via reductive dechlorination. Table 4 shows the compounds detected in the gas phase and the primary biological processes that contribute toward their degradation (i.e., aerobic degradation as a growth substrate, aerobic cometabolic degradation, and anaerobic dechlorination). Some of the organic compounds, such as BTEX, also can be degraded anaerobically, but they are degraded much more rapidly aerobically.

Most of the CACs are degraded fortuitously (i.e., cometabolically) by the action of nonspecific enzymes; those compounds are not used by bacteria as growth substrates for carbon and energy. Others can be used as growth substrates, and a third group falls into both categories. The chlorinated aliphatic compound TCE is a classic example of a compound that degrades only via cometabolism, while VC is an example of a compound that is cometabolically degraded or can serve as a source of carbon and energy for the bacteria. Finally, a fourth group of compounds exists that can only be degraded anaerobically, through a process known as reductive dechlorination. PCE is a classic example of a compound that is only degraded via anaerobic reductive dechlorination. PCE cannot be degraded aerobically as a growth substrate or cometabolically.

This section begins with a discussion of cometabolic degradation of chlorinated ethenes and ethanes, followed by a discussion of the growth-related aerobic degradation of other contaminants in the gas stream. The discussion of cometabolism is grouped by cosubstrate, i.e., methane, toluene and

phenol, ammonia, propene, propane, and isopropylbenzene (IPB [cumene]). The last part of this section describes anaerobic dehalogenation for the halogenated compounds.

TABLE 4. PRIMARY BIOLOGICAL DEGRADATION PROCESSES FOR HALOGENATED AND NONHALOGENATED GAS-PHASE CONTAMINANTS IN THE SVE OFF-GAS WASTESTREAM

Off-gas Organic Compound	Primary Biological Removal Process		
	Aerobic Growth Substrate	Aerobic Cometabolism	Anaerobic Dechlorination
PCE			✓
TCE		✓	✓
c-DCE		✓	✓
1,1-DCE		✓	✓
VC	✓	✓	✓
1,1,1-TCA		✓	✓ a
1,1,2-TCA		✓	✓ a
1,1-DCA	✓	✓	✓ a
1,2-DCB	✓		✓
1,3-DCB	✓		✓
1,4-DCB	✓		✓
Chlorobenzene	✓		
1,2,4-TMB	✓		
1,3,5-TMB	✓		
Methylene Chloride	✓		✓ b
Chloromethane	✓		✓ b
Freon™ 113			✓
Benzene	✓		
Toluene	✓		
Ethylbenzene	✓		
Xylenes, Total	✓		
4-Ethyl Toluene	✓		
Acetone	✓		
Methyl Ethyl Ketone (MEK)	✓		
Methyl Isobutyl Ketone (MIBK)	✓		

[a] May be transformed abiotically under anaerobic conditions.
[b] May be degraded directly under anaerobic conditions.
TMB = trimethylbenzene

B. COMETABOLISM

Bacteria that grow on hydrocarbons typically initiate oxidation by incorporating molecular oxygen into organic compounds by the action of enzymes known as oxygenases (Wackett and Householder, 1989). Two types of oxygenases, monooxygenases and dioxygenases, are involved in the biological oxidation of CACs. The mono- and dioxygenases typically are relatively nonspecific with respect to the types of organic compounds they will attack. The fortuitous oxidation of CACs by bacteria is termed cometabolism.

The oxygenase enzymes do not specifically cleave carbon-halogen bonds, but produce unstable epoxide intermediates that release halides via further chemical or biological decomposition. It is generally accepted that the microorganisms implicated in cometabolism of CACs do not gain energy or carbon for cell growth from the cometabolic oxidation of CACs. In fact, cometabolism often results in the depletion of stored energy reserves in the cell (Alvarez-Cohen and McCarty, 1991a). This implies that an organic cosubstrate other than the CACs is required for biological growth and for the production of the necessary oxygenase enzymes that are used to degrade the CACs. A variety of growth substrates have been used for cometabolic CAC degradation, including methane, aromatic compounds (toluene and phenol), propane, propene, and IPB (cumene). The advantages and disadvantages of each of these cosubstrates will be discussed.

Important issues related to the engineering application of cometabolic CAC degradation include CAC toxicity, competitive inhibition, and intermediate toxicity due to CAC degradation by-products. In particular, many CACs have solvent properties that can adversely affect cell membranes, enzymes, and proteins (Bielefeldt et al., 1995). Because CACs are degraded by the same nonspecific enzymes responsible for the degradation of the growth substrate, competition between the CAC and the growth substrate is known to occur. Competition can reduce the degradation rates of both the growth substrate and the CAC. In addition, multiple CACs can compete with each other (Strand et al., 1990), resulting in reduced CAC degradation rates.

Intermediate toxicity occurs when a by-product of CAC degradation exerts toxic effects on the cells (Alvarez-Cohen and McCarty, 1991a; Bielefeldt et al., 1995). For example, methanotrophs convert TCE to TCE-epoxide (Fox et al., 1990), which can react with and damage intracellular cell protein and DNA (Wackett and Householder, 1989). As the amount of TCE transformation increases, the amount of TCE-epoxide increases and the viability of the cell decreases until it is lost entirely. Intermediate toxicity poses a significant treatment challenge with respect to cometabolic degradation of CACs. Direct TCE toxicity also is a concern; like many organic solvents, TCE is hydrophobic and can express toxic or

inhibitory effects on cells by partitioning onto cellular components, such as lipids and polysaccharides. However, the toxic effects of TCE appear to be much less significant than the effects of intermediate toxicity during TCE degradation.

The following discussion of cometabolic degradation of CACs using different cosubstrates focuses on (1) the ability of the substrate-specific bacteria to degrade different types of CACs (i.e., mono-, di-, and tri- chloroethenes and chloroethanes), (2) competition between the organic cosubstrate and CACs, and (3) intermediate toxicity during CAC biotransformation.

C. OXIDATION OF CHLORINATED ALIPHATIC HYDROCARBONS BY METHANE-OXIDIZING BACTERIA

Wilson and Wilson (1985) were the first to demonstrate the biological cometabolism of TCE, using ^{14}C-labeled TCE; TCE was transformed to CO_2 in a soil column that had been exposed to natural gas with 74% methane. The authors concluded that methanotrophic organisms (the methane-utilizing bacteria) were responsible for the transformation of TCE, and TCE degradation has since been confirmed in numerous other studies using methane-oxidizing bacteria.

Methanogenic bacteria grow aerobically, using methane as a carbon and energy source. The oxygenase enzyme methane monooxygenase (MMO) catalyzes the first step in the biochemical pathway of methane oxidation. MMO requires both oxygen and a high-energy reducing substrate nicotinamide adenine dinucleotide (NADH) to catalyze the oxidation reactions. Formate can be used as an alternative energy source to support intracellular NADH production, but it does not support the production of MMO enzymes, which require methane as the primary substrate (McFarland et al., 1992). In some methanotrophs, MMO is nonspecific and can catalyze the cometabolism of a number of chlorinated aliphatic chemicals, including chloroethenes, chloroethanes, and chloromethanes.

Oldenhuis et al. (1989) showed that two different MMO enzymes are responsible for methane oxidation: Type 1 methanotrophs produce a soluble form of MMO (sMMO), and Type 2 methanotrophs produce a particulate form of MMO (pMMO). TCE was degraded only by cells that expressed sMMO, which demonstrated a much broader substrate range than pMMO. The sMMO was expressed only under low copper concentrations (< 0.25 μM copper [Tsien et al., 1989]); and, when copper was added to the medium, only pMMO was expressed, and no TCE degradation was observed. Oldenhuis et al. (1989) also showed that sMMO catalyzed the degradation of a wide variety of other CACs, including chloroform (CF) and dichloromethane (DCM); 1,1-DCA, 1,2-DCA, and 1,1,1-TCA; 1,1-DCE, *c*-DCE, *t*-DCE, and TCE; 1,2-dichloropropane; and 1,3-dichloropropylene (*t*-1,3-DCP).

Methanotrophic bacteria have been shown to degrade TCE, DCE, and VC at relatively high degradation rates to nondetectable concentrations by a variety of pure and mixed methanotrophic cultures (Oldenhuis, et al., 1989; Alvarez-Cohen and McCarty, 1991b; Tsien et al., 1989; Fogel et al., 1986). The ability to degrade a wide variety of CACs and the ability to rapidly degrade TCE made methanotrophs the subject of further studies about the potential for using them to degrade CACs in the environment.

In spite of the success reported using methanotrophs to cometabolically degrade a wide variety of CACs, including TCE, the application of methanotrophs for the removal of CACs from the environment is restricted by three main factors (Oldenhuis et al., 1989). First, not all methanotrophs can degrade TCE; some organisms, for example, produce exclusively pMMO and as such are unable to degrade TCE. Second, methane inhibits TCE degradation; during methane-supported growth, methane inhibition could limit TCE degradation. Third, TCE degradation intermediates are toxic to methanotrophs.

In a mixed methanotrophic culture, only a fraction of the overall biomass will contribute directly to the degradation of TCE and other CACs, while the entire population will be dependent on methane as a growth substrate. Due to the nature of cometabolism, TCE degradation provides no selective advantage to methanotrophs, and it is improbable that selective enrichment of TCE-degrading methanotrophs under field conditions can be achieved (Oldenhuis et al., 1989). The design and operation of a methanotrophic TCE-degrading culture must be one that enhances sMMO production and stimulates the reducing power (i.e., intracellular NADH production) of the culture, but the presence of other methanotrophs that will compete for methane cannot be avoided. Methanotrophs exhibit first-order TCE degradation rates. First-order degradation rates imply that long treatment periods or high cell concentrations are essential if low final concentrations are required. The fact that only a fraction of the overall biomass will contribute to TCE (or CAC) degradation implies that a large fraction of the biomass will remain unused with respect to CAC degradation.

Because MMO is responsible for both methane oxidation and TCE epoxidation, methane and TCE are considered to be competitive substrates (Alvarez-Cohen and McCarty, 1991a). In the presence of methane, TCE degradation rates are reduced. In column studies using soil microcosms, Lanzarone and McCarty (1990) showed that little or no TCE degradation occurred when the feed methane concentration was 4.5 mg/L, but TCE degradation did occur with a feed concentration of 1.5 mg/L, suggesting competition between methane and TCE for MMO activity. Balancing the drawbacks of enzyme competition and the requirement of methane for growth and maintenance represents a significant

engineering challenge. The kinetics of methane degradation and TCE degradation are such that the MMO enzyme has a much higher affinity for methane than for TCE, so that relatively low methane concentrations can inhibit TCE degradation rates. In the presence of low TCE degradation rates, a large active biomass must be supported to maintain significant TCE biodegradation (Speitel and Leonard, 1992). However, the production of a large biomass culture also requires high methane concentrations, resulting in a cycle of increasing methane concentrations and decreasing TCE degradation kinetics.

The problem of methane and TCE competition has been overcome in the laboratory by using resting cells; cells are grown first on methane, then fed TCE, after the methane is depleted. After the methanotrophs exhaust their ability to degrade TCE via the exhaustion of MMO enzymes and intracellular NADH, they are subjected to another feed cycle, using only methane. Simulating a batch feed cycle in the field is difficult, in part, because methanotrophic bacteria do not always recover from intermediate TCE toxicity after repeated batch feed cycles (Alvarez-Cohen and McCarty, 1991a).

The oxidation of TCE also results in a TCE-epoxide intermediate, which is subsequently degraded to CO_2 and HCl (Little et al., 1988). TCE-epoxide intermediates have been implicated in toxic inhibition of TCE-degrading methanotrophs (Alvarez-Cohen and McCarty, 1991; Henry and Grbi-Gali, 1991; Oldenhuis et al., 1989). The toxicity effects can be permanent, and the methanotrophic bacteria often have difficulty recovering from toxicity caused by TCE-epoxide formation. Even at low concentrations, TCE transformation can result in significant decay of the active biomass, which would be expected to increase with the increasing amounts of transformed TCE (Henry and Grbi-Gali, 1991). In addition, different methanotrophs may be affected by TCE oxidation toxicity to different degrees.

Because of competition and product toxicity, methane degradation and cell growth may not coincide with TCE degradation (Semprini et al., 1990). The development of a two-step system could be required, in which the first step involves growth of methanotrophs under controlled conditions via methane (natural gas) degradation, and the second step involves TCE degradation supported by an alternative energy source for the methanotrophs (i.e., formate, McFarland et al., 1992). Another possibility could be to cycle back and forth between methane growth and TCE degradation, using multiple-sequence batch reactors in parallel with their degradation and growth modes out of phase.

A 2-stage system was investigated by McFarland et al. (1992), where methanotrophs were grown in a separate enrichment reactor and fed into a TCE-degrading reactor. Over 95% TCE degradation (from 29.2 to 1.4 mg/L TCE) was observed, with a volatile solids concentration of 1.3 g/L in the TCE degrading reactor. The mixed methanotrophic culture was ineffective in biodegrading TCE unless it was supplied with formate as an energy source. In the absence of formate as an energy source for NADH, the

removal of TCE occurred mainly through sorption. Unfortunately, the experiment was not conducted over long time periods to demonstrate the long-term ability of methanotrophic cultures to sustain TCE degradation.

Because of the problems described above (first-order degradation rates, competition between methane and TCE, intermediate toxicity), other cosubstrates have been investigated, and their performance has been compared to that of methane.

D. OXIDATION OF CHLORINATED ALIPHATIC HYDROCARBONS BY TOLUENE- AND PHENOL-OXIDIZING BACTERIA

Cometabolic TCE degradation is catalyzed by toluene-oxidizing cultures (Hecht et al., 1995; Mu and Scow, 1994; Wackett and Gibson, 1988) and phenol-oxidizing cultures (Hopkins et al. 1993a; Hopkins et al. 1993b; Folsom and Chapman, 1991; Folsom et al. 1990). Similar to the methane-oxidizing bacteria, which generate the nonspecific enzyme MMO for the initial oxidation step toward methane degradation, the toluene- and phenol-degrading bacteria produce oxygenase enzymes, which are used for the initial oxidation of toluene or phenol degradation, respectively. Those dioxygenase enzymes, like MMO, are relatively nonspecific and have been shown to degrade TCE as well as all three DCEs (Bielefeldt, 1995; Shields and Reagin, 1992; Wackett and Gibson, 1988). However, unlike MMO, they do not appear to be able to degrade chloroethanes (Hopkins et al., 1993b; Shields and Reagin, 1992) or VC and ethylene (Wackett and Gibson, 1988).

Nelson et al. (1988) showed that two strains of *Pseudomonas putida*, Strain PpF1 and Strain B5, utilized a toluene dioxygenase enzyme as the initial step in TCE degradation. Both toluene and phenol induced the production of the toluene dioxygenase enzyme and TCE degradation in both strains. Toluene dioxygenase is not the only enzyme responsible for TCE degradation. *Alcaligenes eutrophus* Strain JMP134 uses a phenol hydroxylase enzyme to degrade TCE (Harker and Kim, 1990), and *Pseudomonas cepacia* G4 (G4) expresses a toluene *ortho*-monooxygenase (TOM) enzyme that has been reported to cometabolically degrade chloroethenes. Thus, a variety of phenol- or toluene-oxidizing bacteria in the environment appear to be capable of TCE oxidation, and different mixtures or ratios of those bacteria may be cultivated in mixed biological cultures used for environmental remediation. *P. cepacia* G4 is probably the most widely studied genus of the aromatic TCE-degrading bacteria. The TOM enzyme is inducible by toluene, phenol, and *o*- or *m*-cresol (Nelson et al., 1986; Nelson et al., 1987).

Both phenol- and toluene-oxidizers appear to have similar characteristics to methanotrophs with regard to product toxicity and competitive inhibition when degrading TCE. However, some phenol degraders have been shown to degrade TCE at higher concentrations without toxic effects, and there have been reports of simultaneous phenol plus TCE degradation (Bielefeldt et al., 1995). Intermediate toxicity was found with *P. cepacia* F1 (Wackett and Householder, 1989) during TCE degradation. TCE oxidation by toluene dioxygenase caused intracellular protein damage, and the cellular toxicity led to reduced growth rates; increasing time of exposure to TCE (from 0 to 9 h) also resulted in increasingly diminished growth rates. However, *P. cepacia* F1 appeared to recover from the toxic effects of TCE oxidation after TCE was removed from the medium.

Bielefeldt et al. (1995) demonstrated that for an inducible, phenol-degrading enrichment, phenol-enhanced TCE degradation rates were as much as twofold higher than endogenous degradation rates. The higher rates continued even after phenol was degraded to nondetectable concentrations. Those results suggest that phenol did not significantly compete with TCE and/or that phenol induced the production of dioxygenase enzymes, resulting in increased TCE degradation. The phenol-degrading culture appeared to be uniquely resistant to TCE intermediate toxicity, endowing the culture with unique engineering advantages based on the fact that higher degradation could be obtained in the presence of phenol. Intermediate toxicity was not observed, and the phenol-degrading culture was able to sustain zero-order TCE degradation rates for 30 h, beginning at a TCE concentration of 25 mg/L.

Mechanistic differences between the dioxygenase and monooxygenase routes of TCE oxidation may account for differences in TCE intermediate toxicity. For *P. cepacia* G4, TCE is degraded by the toluene *ortho*-monooxygenase enzyme, and relatively little apparent toxicity resulted from TCE metabolism (Folsom et al., 1990); the kinetics of TCE metabolism were linear over 3 h of incubation with a supplied TCE concentration of 2.6 mg/L. In contrast, TCE was toxic to *P. putida* F1 (Wackett and Gibson, 1988), which utilizes a toluene dioxygenase enzyme. Decreased TCE degradation rates during the course of TCE metabolism were attributed to the formation of toxic by-products during TCE degradation.

TCE and phenol significantly inhibited each other in phenol-grown *P. cepacia* G4 enrichments (Folsom et al., 1990). TCE inhibited phenol degradation in a concentration-dependent manner. The Michaelis-Menten half-saturation coefficient (K_s) is a measurement of the enzyme affinity for a substrate. Lower K_s values indicate increasing enzyme/substrate affinity. Similarly, K_i is a measurement of the affinity of the enzyme for a competing substrate. The ratio of K_i/K_s can be used to estimate relative levels of inhibition between competing substrates. A ratio of K_s to K_i much less than 1.0 would

suggest a low level of competitive inhibition by the competing compound on the primary substrate degradation rate. A value of 1.0 or higher would suggest a relatively high level of competition. The data-estimated K_s/K_i ratio was measured for *P. cepacia* G4 grown on phenol (Folsom et al., 1990). Competitive inhibition resulted in reduced phenol degradation rates caused by TCE degradation where the K_s/K_i ratio for phenol and TCE was approximately 1, which is consistent with their similar K_s values (Folsom et al., 1990). The apparent K_s for phenol degradation was 0.8 mg/L and the K_s for TCE degradation was approximately 0.4 mg/L. The maximum phenol degradation rate for *P. cepacia* G4 was 111 mg/mg protein-day and for TCE degradation was 1.9 mg/mg protein-day. Because of the rapid phenol degradation kinetics, phenol was degraded to low concentrations when it was the sole carbon source in continuous culture. At phenol concentrations greater than 3.7 mg/L, phenol transiently inhibited its own degradation, indicating that low phenol (and presumably toluene) concentrations must be maintained to prevent substrate toxicity.

Folsom and Chapman (1991) examined the biodegradation of TCE by *P. cepacia* G4 in a bioreactor. *P. cepacia* G4 was grown in a chemostat with phenol, and TCE was degraded in a separate TCE-degrading reactor that was continuously inoculated with the phenol-grown bacteria and fed TCE. Increased phenol concentrations led to increased biomass production and increased TCE degradation rates, with constant specific TCE degradation rates in the side reactor. The maximum potential for TCE degradation was 1.1 mg/mg protein-day and, on average, the reactor degraded 0.7 mg/mg protein-day. The total amount of TCE degraded increased as either reaction time or biomass was increased in the separate TCE-degrading reactor. TCE degradation was observed at concentrations up to 40 mg/L with no significant decreases in rates. At equal concentrations of TCE and phenol in the TCE-degrading reactor, degradation rates were inhibited 50%, consistent with competitive inhibition and similar K_s values determined for TCE and phenol by Folsom et al. (1990).

Hopkins et al. (1993b) demonstrated the use of in situ phenol-degrading microorganisms for TCE degradation. Phenol was fully degraded. The data suggested that competitive inhibition occurred between phenol and *c*-DCE or TCE, and there also were indications that *c*-DCE and *t*-DCE competitively inhibited each other. Cometabolic activity stimulated by phenol addition resulted in higher percent removals of TCE and *c*-DCE than degradation activity stimulated by methane addition, with equal amounts of DO removed. Methane addition resulted in 19 and 43% removal of TCE and *c*-DCE, respectively; phenol addition resulted in 63 and 92% removal, respectively. Removal of TCE per unit of DO consumed for phenol consumption was five times higher than the removal of TCE per unit of DO consumed for methane consumption, indicating that TCE was removed more efficiently by the phenol

oxidizers. Those results could suggest that aromatic compounds are more effective cosubstrates, or they could have been due to site-specific conditions and bacteria.

Heald and Jenkins (1994) showed that TCE oxidation resulted in decreased growth rates and caused rapid cell death, which was attributed to intermediate toxicity of TCE oxidation products. However, TCE induced toluene degradation by whole cells to a rate approximately 40% of that by toluene alone, suggesting that TCE could induce the production of toluene monooxygenase enzymes and presumably its own degradation. TCE could not support growth of the organism. TCE was degraded at concentrations as high as 1.3 mg/L; at higher concentrations, the rate of TCE removal declined rapidly, suggesting direct TCE toxicity to the cells or intermediate toxicity via TCE transformation products.

Hecht et al. (1995) investigated the cometabolic degradation of TCE in a 30-L bubble column bioscrubber, using *P. cepacia* G4 cultures and phenol as the cosubstrate and inducer of toluene dioxygenase enzymes required for TCE degradation. Depending on the gas velocity used, degrees of conversion between 30 and 80% were obtained. Degradation of TCE followed pseudo first-order reaction kinetics. No influence of TCE on the steady-state biomass concentration was detected. The reactor showed good long-term stability, having been operated for several months. Hecht et al. (1995) reported rapid TCE mass transfer, and modeling results showed that the process was mainly limited by reaction rate rather than by the mass transfer rate of TCE. Thus, the best way to increase reactor efficiency would be the use of microorganisms with considerably higher degradation rates to overcome the reaction rate limitations, increased reactor volumes, or increased biomass concentrations.

E. OXIDATION OF CHLORINATED ALIPHATIC HYDROCARBONS BY AMMONIA-OXIDIZING BACTERIA

Arciero et al. (1989) were the first to demonstrate that ammonia-oxidizing bacteria could catalyze the degradation of TCE. Suspensions of *Nitrosomonas europaea* were shown to catalyze the complete disappearance of TCE. However, ammonia oxidation was inhibited 98% in the presence of 1.1 mM TCE. Complete ammonia-oxidizing activity was recovered once TCE was completely degraded. TCE degraded at an initial rate of at least 0.21 mg/mg protein-day. With aged cells, the addition of ammonia appeared to stimulate the rate of TCE degradation.

Three classes of CAC toxicity have been identified according to their inactivating potential (Rasche et al., 1991): (I) compounds that are not biodegraded by *N. europaea* and that have no toxic effect on the cells, (II) compounds that are cooxidized by *N. europaea* and that have little or no toxic

effect on the cells, and (III) compounds that are cooxidized and that have significant intermediate toxicity effects on ammonia oxidation by *N. europaea*. Carbon tetrachloride (CT) and PCE, neither of which are capable of being aerobically biodegraded, fall under Class I, suggesting that the solvent toxicity is not caused by direct cell damage by the solvent, but by intermediate toxicity via solvent degradation products. Chloromethane, chloroethene, and 1,2-DCA fall under Class II. Class III compounds include DCM and CF; 1,1-DCA, 1,1,1-TCA, 1,1,2-TCA, and (1,1,2,2-Tetrachloroethane [TeCA]; VC, 1,1-, *c*- and *t*-DCE, and TCE.

Vannelli et al. (1990) also showed that CT and PCE were not degraded by *N. europaea*, but that dibromomethane, vinyl bromide, and *cis*- and *trans*-dibromoethylene were degraded by *N. europaea*. Based on these results and those reported by Rasche et al. (1991), *N. europaea* has a very broad CAC substrate range, similar to methanotrophs.

TCE is a potent competitive inhibitor of ammonia oxidation by *N. europaea* (Hyman et al., 1995). The K_i value for TCE (30 μM) was similar to the K_s value for ammonia (40 μM), and very low ammonia concentrations inhibited TCE degradation. Increasing TCE concentrations led to increasing levels of ammonia inhibition.

F. OXIDATION OF CHLORINATED ALIPHATIC HYDROCARBONS BY PROPANE-, PROPENE-, AND ISOPROPYLBENZENE-OXIDIZING BACTERIA

Methanotrophs, toluene- and phenol-oxidizing bacteria, and ammonia-oxidizing bacteria represent three groups bacteria that cometabolically degrade TCE and other CACs. Bacteria in the environment oxidize a wide variety of other natural and human-made hydrocarbons. Typically, bacteria initiate their oxidation to grow on hydrocarbons by incorporating oxygen from the atmosphere into organic compounds through the action of enzymes known as oxygenases (Wackett et al., 1989). The oxygenases are generally divided into two groups, the monooxygenases and the dioxygenases. The unique ability of mono- and dioxygenase enzymes to degrade TCE is attributed to their relative nonspecificity. Many other cosubstrates support bacteria that produce oxygenases capable of TCE degradation, including propane (Wackett et al., 1989; Malachowsky et al., 1994), propylene (Ensign et al., 1992; Reij et al., 1995b), and IPB (Dabrock et al., 1992).

Wackett et al. (1989) examined the ability of microorganisms that degrade propane, hexane, cyclohexane, preocene, and nitropropane to degrade TCE. Of the 14 organisms examined, only the 5 propane oxidizers degraded TCE. Those results suggest that TCE oxidation and subsequent biodegradation are not necessarily a general property of oxygenase enzymes and bacteria that contain

those enzymes. The propane-oxidizing bacterium *Mycobacterium vaccae* JOB5 degraded all three DCEs and VC, but not PCE. The order of degradation rates were: VC › *c*-DCE › 1,1-DCE › TCE › *t*-DCE.

TCE degradation by two propane-oxidizing *Rhodococcus* species was investigated by Malachowsky et al. (1994). The *Rhodococcus* species degraded TCE and VC with propane as a growth cosubstrate. Cell suspensions degraded TCE at concentrations up to 5 mg/L and VC at concentrations up to 20 mg/L. Propane competitively inhibited TCE degradation; cell suspensions containing 1 mg/L TCE and 40% propane in the headspace degraded 23% of the TCE, compared to 87% TCE degradation without propane. Cell suspensions degraded up to 5 mg/L of TCE and up to 40 mg/L of VC. Propane-grown resting cell suspensions also degraded chloroform (CF), 1,1-DCE, *c*-DCE, and 1,1,1- and 1,1,2-TCA, suggesting that propane-oxidizing bacteria can degrade a wide variety of different CACs, similar to methanotrophs and ammonia-oxidizing bacteria. PCE, CT, and *t*-DCE were not degraded by the propane oxidizers. The *Rhodococcus* isolates also degraded benzene, toluene, sodium benzoate, naphthalene, biphenyl, and *n*-alkanes ranging in size from propane to hexadecane as carbon and energy sources.

Propylene also can serve as a suitable cosubstrate for TCE degradation. Propylene-grown *Xanthobacter* Py2 cells degrade TCE, but the transformation capacity may be limited and depends on both the TCE and the biomass concentrations (Reij et al., 1995b) . The affinity of Strain Py2 for TCE was low (i.e., a high K_s), which allowed it to grow on propylene while degrading TCE, but also resulted in competition between propylene and TCE and thus in reduced TCE degradation rates in the presence of propylene. TCE degradation was inhibited by high propylene concentrations and did not start before most of the propylene had been consumed. Propylene degradation rates also were inhibited by the presence of TCE. In spite of the competitive inhibition of propylene on TCE degradation, low propylene concentrations can stimulate CAC degradation, although at high concentrations propylene becomes extremely inhibitory (Ensign et al., 1992).

Propylene-grown *Xanthobacter* strains degrade TCE, VC, *c*- and *t*-DCE, 1,3-DCP, and 2,3-DCP, whereas 1,1-DCE and PCE are not degraded (Ensign et al., 1992). *Xanthobacter* is subject to some of the same toxicity problems as described for methane-, toluene-, phenol-, ammonia-, and propane-oxidizing bacteria. However, Reij et al. (1995b) reported that biomass yields on propylene were not affected by the cometabolic degradation of TCE, suggesting that TCE degradation was not toxic to the *Xanthobacter* propylene degraders.

Bacteria grown on IPB also demonstrated CAC degradation abilities (Dabrock et al., 1992). IPB-, phenol-, toluene-, citronellol-, and dipentene-grown cells were tested for their ability to

cometabolically degrade TCE; 27 out of 27 IPB-grown strains tested positive for TCE degradation, compared to 13 out of 27 toluene-degrading strains, 2 out of 9 phenol-degrading strains, and zero out of 4 citronellol- and 11 dipentene-degrading strains, respectively, indicating that a high proportion of IPB-degrading bacteria can degrade TCE. *Rhodococcus erythropolis* BD1 degrades TCE and *c*- and *t*-DCE with IPB as a primary growth substrate, whereas *Pseudomonas* species Strain JR1 degrades all three DCEs, VC, 1,1,2-TCA, and 1,2-DCA (Dabrock et al., 1992). Dabrock et al. (1992) showed that TCE degradation rates with IPB-oxidizing bacteria increased with increasing initial TCE concentrations, and a linear dependence was observed between 0 and 200 μM. The time after which the activity ceased appeared to be concentration-independent, and the apparent inactivation did not appear to be a result of toxic TCE effects because it was overcome by substrate refeeding. Thus, similar to some phenol-oxidizing cultures described above, the IPB-oxidizing bacteria appear to be resistant to intermediate TCE toxicity. The maximum initial TCE degradation rate for *R. erythropolis* BD1 was 1.2 mg/mg protein-day, which was comparable to the TCE degradation rate for *P. cepacia* G4 (Folsom et al., 1990).

G. SUMMARY OF COMETABOLISM

Several cosubstrates have been implicated in the cometabolic degradation of CACs. They include methane, ammonia, propane, propylene, and aromatic compounds such as toluene, phenol, and IPB. Table 5 shows advantages and disadvantages of each cosubstrate. A common disadvantage of all the cosubstrates is the intermediate toxicity that occurs during TCE oxidation. However, some phenol- and IPB-degrading bacteria appear to be able to degrade TCE at higher concentrations without toxic effects. Bielefeldt et al. (1995) reported simultaneous phenol plus TCE degradation without intermediate toxicity, and relatively little apparent toxicity results from TCE metabolism by *P. cepacia* G4 (Folsom et al., 1990). Dabrock et al. (1992) reported that the IPB degraders also were not affected by intermediate toxicity; however, no data were presented supporting their argument. In contrast, there are no reports of methanotrophs, ammonia oxidizers, or propane oxidizers that are resistant to intermediate toxicity. Another common disadvantage of cometabolic CAC degradation is competitive inhibition between the cosubstrate and TCE (and presumably among the CACs themselves). Competitive inhibition has been reported for nearly all of the cosubstrates investigated. However, competition results in different degrees of inhibition of TCE degradation, depending on the species of bacteria and its relative affinities for TCE and the cosubstrate. For example, the ammonia oxidizers appear to be among the most severely inhibited bacteria with respect to TCE degradation, where only very low ammonia concentrations can result in

significantly reduced TCE degradation rates. Methane, phenol, and toluene inhibit TCE degradation to varying degrees.

An important feature of the methanotrophs, the ammonia oxidizers, and the propane oxidizers is their ability to degrade chloroethanes in addition to TCE and other chloroethenes. Of the aromatic-degrading bacteria, only the IPB degraders were reported to degrade both chloroethenes and chloroethanes. The phenol- and toluene-degrading bacteria repeatedly have been shown not to degrade chloroethanes.

Other important features of the different cosubstrates include: whether they are explosive, their aqueous solubilities, the body of literature supporting their use, the proportion of cosubstrate-oxidizing bacteria that directly contribute toward CAC degradation, and bulk costs. Methane and propane are explosive and have very low aqueous solubilities, which would necessitate their application at high gas-phase concentrations. Phenol, toluene, and IPB are not explosive and have much higher aqueous solubilities, but they are more toxic to bacteria at higher concentrations than the other cosubstrates, and they require more careful process control to ensure that they are completely degraded. The number of cosubstrate-oxidizers that contribute toward CAC degradation will impact the cost of their application. For example, if a low proportion of methanotrophs in a methane-fed reactor degrades TCE, than a large fraction of the methane added to the system will not contribute toward TCE degradation, resulting in inefficient methane use. Dabrock et al. (1992) investigated 27 different IPB-degrading strains of bacteria, and all 27 degraded TCE. Only 13 out of 27 toluene-degraders and 2 out of 9 phenol-degraders degraded TCE. These results suggest that not all of the toluene- and phenol-degrading bacteria can degrade TCE, but a high percentage (possibly 100%) of IPB-degrading bacteria appear to degrade TCE. That would make IPB a relatively efficient cosubstrate for TCE degradation.

Another common disadvantage of cometabolic CAC degradation is competitive inhibition between the cosubstrate and TCE (and presumably among the CACs themselves). Competitive inhibition has been reported for nearly all of the cosubstrates investigated. However, competition results in different degrees of inhibition of TCE degradation, depending on the species of bacteria and its relative affinities for TCE and the cosubstrate. For example, the ammonia oxidizers appear to be among the most severely inhibited bacteria with respect to TCE degradation, where only very low ammonia concentrations can result in significantly reduced TCE degradation rates. Methane, phenol, and toluene inhibit TCE degradation to varying degrees.

TABLE 5. ADVANTAGES AND DISADVANTAGES OF DIFFERENT COSUBSTRATE-OXIDIZING BACTERIA FOR COMETABOLIC CAC DEGRADATION

Cosubstrate	Advantages	Disadvantages
Methane	Methanotrophs degrade a wide variety of CACs Formate can supply additional energy Extensive body of literature available	Competitive inhibition Intermediate toxicity Requires low aqueous Cu^{2+} concentrations Low solubility Explosive Low proportion of CH_4-oxidizers contribute to CAC degradation
Toluene/Phenol	High solubility Rapid growth Simple process control Concurrent phenol- and CAC-oxidation is possible Selected cultures do not exhibit intermediate toxicity, and those strains may be resistant to intermediate toxicity Extensive body of literature available	Phenol-oxidizers do not degrade chloroethanes Competitive inhibition Low proportion of phenol-oxidizers contribute to CAC degradation
Ammonia	Ammonia-oxidizers degrade a wide variety of CACs Cells can recover from intermediate toxicity in the absence of CACs High solubility Inexpensive	Severe competitive inhibition Intermediate toxicity Produces nitrate Limited literature information available Low proportion of ammonia-oxidizers contribute to CAC degradation
Propane	Possibly degrade a wide variety of CACs	Low solubility Explosive Competitive inhibition Intermediate toxicity Limited body of literature available
Propylene	High solubility Low levels of intermediate toxicity	Propylene-oxidizers do not degrade chloroethanes Competitive inhibition Limited body of literature available
Isopropylbenzene [IPB (cumene)]	Degrade a wide variety of CACs High solubility Simple process control Possibly low competitive inhibition High percentage of IPB-oxidizers may degrade CACs Some strains may be resistant to intermediate toxicity	Low solubility Explosive Competitive inhibition Intermediate toxicity Limited body of literature available

Table 6 summarizes specific TCE degradation rates and concentration tolerances of cosubstrate-degrading enrichments (modified from Bielefeldt et al., 1995) for comparison. It is difficult to make a specific comparison of all the cultures because of their different growth and environmental conditions. In addition, pure cultures are expected to have much higher specific growth rates than mixed cultures because a much higher percentage of the normalized biomass concentration contributes directly to TCE degradation in pure cultures. In general, the phenol-degrading enrichments reported the highest endogenous transformation capacities (Tc), which is a measurement of the observed mass of TCE degraded per mass of protein inactivated by TCE degradation. The degradation rates can be used to determine relative differences in the abilities of different cosubstrate-degrading bacteria to degrade CACs.

The highest concentration degraded, shown in Table 7, more frequently represents the maximum TCE concentration tested, rather than a toxicity threshold. The fact that the tested TCE concentrations often exceeded the projected total aqueous CAC concentration from the SVE off-gas suggests that CACs will not be present at toxic concentrations in the reactor(s).

Table 7 shows the cosubstrate transformation capacities of TCE using different cosubstrate-oxidizing cultures, measured as the cosubstrate consumed per mass of TCE degraded. A wide range of transformation capacities has been reported, suggesting that the amount of cosubstrate required for CAC degradation is culture-specific, and it may depend upon specific experimental and environmental conditions. Thus, the optimal amount of cosubstrate required for CAC degradation in the SVE off-gas will have to be determined experimentally in the laboratory and field demonstrations. For a steady-state system, optimal cosubstrate concentrations must be controlled so that they can support a cosubstrate-degrading population large enough to degrade the CACs to predetermined effluent concentrations, but low enough to limit or prevent competitive inhibition between the cosubstrate and the CACs. Lower cosubstrate concentrations also will result in reduced cosubstrate costs during full-scale operation.

TABLE 6. REPORTED MAXIMUM TCE DEGRADATION RATES AND TCE CONCENTRATIONS TESTED FOR DIFFERENT COSUBSTRATE-OXIDIZING CULTURES

Culture	Cosubstrate	Reported TCE degradation rate (g/g VSS-d)	Highest TCE concentration degraded/tested (mg/L)	Test Temp (°C)	Reference
OB3b	Methane	1.58-3.78	10.6	30	Tsien et al. (1989)
Mixed methanotroph	Methane	0.58 - 2.0	21	21	Alvarez-Cohen and McCarty (1991a)
Mixed methanotroph	Methane	0.015	-		Phelps et al. (1990)
Mixed methanotroph	Methane	0.021-0.026	15	28	McFarland et al. (1992)
Mixed methanotroph	Methane	0.027	7.8	20	Strand et al. (1990)
P. putida F39/D	Phenol	0.62	› 9.4	30	Zylstra et al. (1989)
P. putida	Toluene	1.2	1.6	30	Heald and Jenkins (1994)
P. cepacia G4	Phenol		» 70, ‹ 428	23	Shields and Reagin (1992)
P. cepacia G4 PR1	Phenol	0.09	› 70, ‹ 428	23	Shields and Reagin (1992)
P. cepacia G4	Phenol	1.1 (0.7)	50	28	Folsom and Chapman (1991)
P. cepacia G4	Phenol	1.92	30	26	Folsom et al. (1990)
P. cepacia G4	Toluene	0.12-0.18	-	28	Landa et al. (1994)
Filamentous enrichment	Phenol	0.10-0.25 (0.18)	130	20	Bielefeldt et al. (1995)
A. eutrophus JMP134	Phenol	0.05	-	30	Harker and Kim (1990)
N. europaea	Ammonia	0.21	1.4	20	Arciero et al. (1989)
M. vaccae JOB5	Propane	0.07	4.15	--	Wackett et al. (1989)
Xanthobacter PY2	Propylene	0.36	41.5	30	Reij et al. (1995b)
R. erythropolis BD1	Isopropylbenzene IPB (cumene)	1.2	33	20	Dabrock et al. (1992)

TABLE 7. REPORTED TCE TRANSFORMATION CAPACITIES OF DIFFERENT COSUBSTRATES

Culture	Cosubstrate	Transformation Capacity (g cosubstrate consumed per g of TCE degraded)	Reference
Soil microcosm	Methane	1,000	Hopkins et al. (1993a)
M. trichosporium Ob3b	Methane	320-1,200	Oldenhuis (1992)[a]
Mixed culture	Methane	77	Alvarez-Cohen and McCarty (1991a)
Mixed culture	Methane	11-30	Phelps et al. (1990)
Soil microcosm	Phenol	9	Hopkins et al. (1993a)
	Phenol	50	Folsom and Chapman (1991)
Soil, in situ	Phenol	500	Hopkins et al. (1993b)
P. cepacia G4	Toluene	14-71	Landa et al. (1994)
P. cepacia G4	Phenol	2-10	Ensley (1992)
P. cepacia G4	Toluene	4-40	Ensley (1992)
N. europaea	Ammonia	1.6	Arciero et al. (1989)
Xanthobacter PY2	Propylene	4-23	Reij et al. (1995b)

[a] As cited in Reij et al., 1995b.

H. DIRECT CAC DEGRADATION

Most bacteria capable of cometabolic TCE degradation also are able to cometabolically degrade DCEs and VC. However, VC appears to be unique among the chloroethenes in its ability to be degraded as a sole carbon and energy source (Hartmans and de Bont, 1992). *Mycobacterium aurum* L1 grows on VC as a sole carbon and energy source. Hartmans and de Bont reported that three additional strains similar to Strain L1 and identified as *M. aurum* grew on VC as a sole carbon and energy source. The rate of VC degradation was 5 mg/mg cells-day. *M. aurum* L1 also oxidized dichloroethenes at rates that were in the same range as VC; 1,1-DCE was degraded at 1.4 mg/mg cells-day, *c*-DCE was degraded at 4.2 mg/mg cells-day, and *t*-DCE was degraded at 3.5 mg/mg cells-day. The authors did not report whether the DCE degradation contributed carbon and energy toward cell growth, and TCE was not degraded.

Several organisms have been shown to grow on 1,2-DCA as a sole carbon and energy source (Janssen et al., 1985; Strotmann et al., 1990; van der Ploeg et al., 1994). However, similar to chloroethenes, the higher chlorinated ethanes are degraded only cometabolically under aerobic

conditions. Organisms capable of utilizing CACs for growth can be used for the removal of these substrates from wastestreams such as the contaminated SVE off-gas at McClellan AFB.

I. DEGRADATION OF ADDITIONAL CONSTITUENTS: DICHLOROBENZENES, BTEX, AND ACETONE

Pure cultures of *Pseudomonas* (Haigler et al., 1988; Spain and Nishino, 1987) and *Alcaligenes* (de Bont et al., 1986; Schraa et al., 1986) and *Xanthobacter* strains (Spiess et al., 1995), which use DCBs as the sole source of carbon and energy, have been shown. Each species appears to attack DCBs by a dioxygenase, producing chlorocatechols that are subsequently degraded. The initial oxidation of DCBs appears to be the rate limiting step toward their degradation (Schraa et al., 1986); and growth rates are generally slow, requiring large bioreactor volumes or long solids retention times. Long solids retention times can be provided by a fixed-film biological process, which retains biomass longer than complete-mix systems.

In general, BTEX compounds and acetone readily degrade in aerobic biological systems. Their degradation rates are very fast, and they can be degraded efficiently to relatively low concentrations in biological systems (Wiedemeier et al., 1995; Bielefeldt et al., 1995 (BTEX only)). Because BTEX and acetone are easily and rapidly degraded, they are expected to be among the first compounds to be degraded in the contaminated off-gas stream, followed by increasingly recalcitrant compounds.

J. ANAEROBIC REDUCTIVE DECHLORINATION

Many of the halogenated compounds discussed above are relatively recalcitrant to aerobic degradation. As a general rule, increasing the number of chlorines on a carbon-based molecule renders it more difficult to degrade aerobically. The opposite can be said under anaerobic conditions. Under anaerobic conditions, the more-chlorinated compounds tend to be dechlorinated more rapidly than the less-chlorinated compounds, possibly because they are in a more oxidized state due to the chlorine halogens. Thus, the order of chloroethene degradation rates is PCE › TCE › DCE » VC. Usually, VC is the rate-limiting step for complete PCE dechlorination to ethene, and it tends to accumulate in the environment or in biological systems. Similarly, higher-chlorinated benzene compounds tend to be more easily dechlorinated under anaerobic conditions than the lower chlorinated benzenes, such as chlorobenzene and most DCBs.

Anaerobic dechlorination has been well established for chloroethenes and chloroaromatic compounds. Less information is available for chloroethanes, although they too can be dechlorinated anaerobically. The chloroethenes are dechlorinated via a relatively simple pathway that involves the step-wise removal of chlorine to form ethene as the final product. Unlike the chloroethenes, chloroethanes can be dehalogenated to lower chloroethanes or they can be degraded biologically or abiotically to lower chlorinated ethenes (Chen et al., 1996; Vogel et al., 1987), which involves the transformation of the carbon-carbon bond to a double bond during dechlorination. 1,1,1-TCA transformations to 1,1-DCA (Galli and McCarty, 1989) and 1,1-DCE have been reported (Vogel et al., 1987), and 1,1,2-TCA can be transformed to 1,2-DCA or VC (Chen et al., 1996).

Enzien et al. (1994) reported the reductive dechlorination of PCE and TCE under bulk aerobic conditions in a sediment column. The reductive dechlorination was attributed to micro anaerobic communities in the aerobic sediment column, suggesting the potential for simultaneous aerobic and anaerobic biotransformation processes under bulk aerobic conditions. Methane production also contributed to the conclusion that anaerobic activity was present in the sediment column. PCE and TCE were dechlorinated to *c*-DCE which showed no sign of being dechlorinated to VC.

SECTION IV

VAPOR-PHASE BIOLOGICAL REACTOR TECHNOLOGY

Gas-phase biotreatment of contaminated vapors containing toxic air contaminants (TAC) has gained interest over the last two decades with the discovery of biological cultures that can degrade those contaminants. Traditional vapor scrubbing, thermal incineration, catalytic incineration, and adsorption to activated carbon have all been used to treat airborne contaminants in the past. However, all these methods are potentially more expensive than biotreatment (Chetty et al., 1992; Dharmavaram, 1991). In addition to economic issues, another drawback of both traditional vapor scrubbing and adsorption to activated carbon is that these methods do not destroy the toxic contaminants of interest, but merely transfer them from one medium (air) to another (i.e., liquid or solid) medium. Further processing is necessary to destroy the contaminants. Biotreatment processes are environmentally friendly, and they produce only nonhazardous byproducts such as additional biomass, water, and low levels of carbon dioxide. No carbon monoxide, NO_x, Sulfur oxides (SO_x), or thermal pollution is produced.

A. CONVENTIONAL BIOFILTERS

One type of biological air treatment process commercially available is biofiltration (Leson and Winer, 1991; van Groenestijn and Hesselink, 1993; VDI Richtlinie 3477, 1991). Biofiltration is a process that utilizes microorganisms immobilized in the form of a biofilm layer on a porous, absorbent filter packing material. As a contaminated vapor stream passes through the filter bed, pollutants are transferred from the vapor to the biolayer and are oxidized, forming carbon dioxide and water, or, in the case of odors, are transformed into less- or nonodorous compounds. The packing material used in conventional biofilters is usually composed of compost, wood chips, peat, heather, or combinations of these materials (Leson and Winer, 1991; VDI Richtlinie 3477, 1991). In addition, bulking agents, such as polystyrene beads, and pH buffering agents such as calcium carbonate or lime, can be added to enhance the mechanical and performance properties of the packing (van Lith et al., 1990). Activated carbon has also been investigated as a packing material for petroleum hydrocarbon and ethanol applications (Hodge et al., 1991; Hodge and Devinny, 1994; Devinny and Hodge, 1995).

The simplest form of biofiltration system is the soil bed, where a horizontal network of perforated pipe is placed 2 to 3 feet below the ground (Kampbell et al., 1987; Bohn and Bohn, 1988; Bohn, 1992). Vapor contaminants are pumped through the piping, flow upward through the soil pores,

and are oxidized by microorganisms present within the soil. However, efficient and reliable biofiltration requires a much more controlled environment than typically found within soil beds. Control of temperature, bed moisture content, and pH is necessary if the microorganisms responsible for biodegradation are to function efficiently. The need for a controlled environment has led to the development of sophisticated units containing better defined filter support and organisms cultured within the laboratory (van Lith et al., 1990; Togna et al., 1993). Biofilters are most economical for treating high-volume air flows (1,000 to 100,000 cfm and above) containing low concentrations (less than 1,000 ppmv) of biodegradable organic compounds (Dyer and Mulholland, 1994; Vembu and Walker, 1995).

To prevent the biofilter bed from drying out, the influent vapor stream usually is prehumidified to as close to 100% water saturation as possible. Water typically is added directly to the packing material to replace water lost from the packing due to the exothermic nature of the biological oxidation process (van Lith et al., 1990). Both upflow (countercurrent) and downflow (cocurrent) configurations have been used.

Biofiltration has been used in Europe for over 30 years to control odorous air emissions (Leson and Winer, 1991). Biofilters have also been used in the United States to treat hydrogen sulfide, mercaptans, alcohols, and other odor-causing airborne contaminants emitted from wastewater treatment plants, industrial process streams, and composting facilities (Allen and Yang, 1992; Yang and Allen, 1994a; Yang and Allen, 1994b; Leson et al., 1993; Kuter et al., 1993). Recent developments in biofilter technology have expanded the range of treatable target compounds to include a wide range of organic contaminant air pollutants (Leson and Winer, 1991; Fouhy, 1992; Togna et al., 1993; Ergas et al., 1993; Yavorsky, 1993; Langseth and Pflum, 1994; Lacky and Holt, 1996). One such advance has been the development of biofilters to treat petroleum hydrocarbon vapors (Peters et al., 1993; Zurlinden et al., 1994; Hodge et al., 1991; Apel et al., 1993; Togna et al., 1994; Leson and Smith, 1995). In cases where off-gases are produced during vapor extraction operations containing low to moderate concentrations of hydrocarbons, biofiltration has the potential to be a cost-effective alternative to conventional air treatment technologies such as incineration and carbon adsorption.

Biofiltration systems have been investigated for the removal of TCE and other chlorinated compounds from wastewater treatment plant emissions (Ergas et al., 1992; Webster et al., 1995). Typically, these compounds are present at very low concentrations (100 ppbv) within these streams (Ergas et al., 1992). For these applications, the biofilter systems usually rely on trace levels of aromatic compounds or methane in the vapor stream to act as cosubstrates for TCE removal. TCE DREs typically are only 50% at economically viable reactor vapor contact times (Ergas et al., 1992; Webster et al.,

1995), where the vapor contact time is defined as the reactor volume occupied by packing divided by the vapor flowrate.

Biofilters have also been investigated for removal of TCE at higher concentrations (10 to 50 ppmv) using methane or propane as cosubstrates (Griffiths et al., 1995; HazTech News, 1996). In one test, 80% TCE removal could be attained for a short period of time (9 days) after pulsing with propane, but only at a very long vapor contact time of 55 minutes (Griffiths et al., 1995). During normal operation, at a steady propane or methane feed, the TCE removal efficiency was approximately 50%. In another test, greater than 90% TCE removal could be attained at a 30-minute vapor contact time (HazTech News, 1996). This test also used propane as the cosubstrate.

B. FIXED-FILM TRICKLING FILTERS (BIOTRICKLING FILTERS)

A second type of biological system used to treat gas-phase organic contaminants is the biotrickling filter. Biotrickling filters are similar to biofilters, but contain a stable, solid packing material instead of compost or peat, and operate with liquid medium flow over the packing to facilitate mass transfer (Dharmavaram, 1991; Hartmans and Tramper, 1991; Togna and Folsom, 1992; van Groenestijn and Hesselink, 1993; Togna and Singh, 1994). Only the recirculating liquid is inoculated initially with microorganisms, but a biofilm layer forms on the packing shortly after startup. Contaminants are transferred to, and degraded by, microorganisms present within both the recirculating liquid and the biofilm layer, although the majority of the degradation is performed within the biofilm. Biotrickling filters can be operated in either an upflow mode, countercurrent to the flow of the recirculating medium, or in a downflow mode, cocurrent to the flow of the recirculating medium. However, the downflow mode of operation has been shown through mathematical modeling to result in higher removal efficiencies for less water-soluble contaminants because absorbed contaminants will not be restripped from the recirculating water during this mode of operation (Ockeloen et al., 1992). Previously, it had been thought that biotrickling filters were not capable of effectively treating sparingly soluble contaminants with Henry's law coefficients greater than 0.02 atm-m^3/mol (van Groenestijn and Hesselink, 1993). However, it has been shown recently that biotrickling filters are more effective than biofilters for treatment of isopentane, which has a Henry's law coefficient of 1.3 atm-m^3/mol, possibly due to the adsorptive influence of the microbial biomass (Togna and Singh, 1994).

Biotrickling filters have some distinct process advantages over conventional biofilters. First, the pH of the recirculating liquid within biotrickling filters is easily monitored and controlled by the automatic addition of acid or base. The pH within conventional biofilters is controlled by the addition of

solid calcium carbonate or lime to the packing material at the beginning of operation (van Lith et al., 1990). Once the buffering capacity is exhausted, and the pH of the bed material drops, the filter bed is removed and replaced with fresh material. For the treatment of air streams containing halogenated or sulfur-containing contaminants that generate acids upon biodegradation, biofilter bed replacement can be quite frequent (Ergas et al., 1995). Therefore, biotrickling filters are frequently more cost effective than biofilters for treating halogenated contaminants such as TCE and methylene chloride, or sulfur compounds such as hydrogen sulfide (H_2S) and carbon disulfide (CS_2) (Hartmans and Tramper, 1991; Diks and Ottengraf, 1991a; Diks and Ottengraf, 1991b; van Lith et al., 1993). However, in cases where only H_2S and/or CS_2 are/is present, biofilters have been shown to be capable of sustained contaminant removal under low pH conditions (Yang et al., 1993a; Yang et al., 1993b; Yang et al., 1994; Yang and Alibeckoff, 1995). Second, biotrickling filters allow for much greater control of process parameters, including salt removal and nutrient/supplemental food addition, and therefore are much more flexible and durable than biofilters. This is especially important for treatment of air streams containing recalcitrant contaminants such as TCE that can be treated only by adding a cosubstrate to the system. Both biofilters and biotrickling filters can be used to treat air streams containing TCE using methanotrophs by feeding methane to the air stream. However, liquid cosubstrates (such as phenol and toluene) and sources of reducing equivalents (such as formate) are more easily added to biotrickling filters than to biofilters. Finally, biotrickling filters may offer higher removal efficiencies at higher organic loadings than biofilters, possibly due to thicker biofilm development or the adsorptive influence of the microbial biofilm (Togna and Folsom, 1992; Togna and Singh, 1994).

Biotrickling filters also offer space advantages compared to conventional biofilters. The packing height in conventional biofilters is usually limited to 4 feet due to problems of bed compaction over time. Therefore, biofilters tend to occupy a significant area. Biotrickling filters, however, often show significantly better performance than biofilters (Togna and Folsom, 1992; Togna and Singh, 1994), and therefore can be designed as smaller skid-mounted units, allowing for much more efficient use of space. In addition, biotrickling filters contain inert packing materials that are not subject to deterioration and compaction. Therefore, packing replacement is not required, and the systems can be designed as columns. Packing materials typically used within biotrickling filters include corrugated plastic packings (Togna et al., 1995; Torres-Cardona et al., 1993), dumped plastic and metal scrubber packing (Hartmans and Tramper, 1991; Pedersen and Arvin, 1995), carbon-impregnated polyurethane foam (DeFilippi et al., 1993), structured ceramic packing (Govind et al., 1993), activated carbon (Govind et al., 1993), Celite® (Sorial et al., 1994; Sorial et al., 1995), and even oyster shells (Shields et al., 1993).

Considerable attention has been devoted recently to biomass growth-control mechanisms within biotrickling filters. These mechanisms primarily involve operational/biological control methods. Three general control methods have been investigated. These are (1) limiting an inorganic nutrient such as nitrogen (Weber and Hartmans, 1994) or potassium (Holubar et al., 1995), (2) increasing the ionic strength (Weber and Hartmans, 1994) or salt (NaCl) concentration (van Lith et al., 1994; Diks et al., 1994a), and (3) incorporating mechanical liquid/vapor shear mechanisms (Togna et al., 1995) or backwashing (Sorial et al., 1994; Sorial et al., 1995). However, nitrogen limitations have been shown to significantly decrease bioreactor performance (Holubar et al., 1995). Diks et al. (1994b) recently established a steady-state biomass concentration within a biotrickling filter treating methylene chloride, where almost all of the methylene chloride carbon was converted to carbon dioxide. This steady-state biomass concentration in the biotrickling filter was attributed to rapid endogenous respiration and/or predation by protozoa that countered the growth of methylene chloride degrading bacteria (Diks et al., 1994b).

Recent field-pilot and full-scale biotrickling filter applications include treatment of (1) methylene chloride emissions from an artificial glass production process (van Lith et al., 1993); (2) N, N-dimethylacetamide emissions released during production of Lycra® fibers (Dharmavaram et al., 1995); (3) acetone and ethanol emissions from a cosmetic pencil production process (Loy, 1995); (4) ethanol and ethyl acetate emissions from a printing operation (Loy, 1995); (5) odorous vapor emissions released during the production of tobacco and compost (Loy, 1995); (6) hydrogen sulfide and carbon disulfide emissions from cellophane, rayon, and sponge production processes (Revah et al., 1994; Revah et al., 1995); (7) ammonia emissions from a composting operation (Smits et al., 1995); (8) isopentane and isobutane emissions from a foam manufacturing facility (Togna et al., 1995); and (9) wastewater treatment plant emissions, including low concentrations of chlorinated organic contaminants, by Envirogen, Inc.

Biotrickling filter systems have been investigated for removal of TCE from air streams (Speitel and McClay, 1993; Shields et al., 1993; Govind et al., 1993; Govind et al., 1995). Speitel and McClay (1993) demonstrated 20 to 80% removal of TCE at packed-bed vapor contact times of 5 to 12 min using methane as a cosubstrate and the bacterium *Methylosinus trichosporium* OB3b. The TCE concentration range investigated by Speitel and McClay was between 300 and 1,000 μg/L (55 and 190 ppmv). Shields et al. (1993) investigated removal of TCE from air using a constitutive mutant of *P. cepacia* G4. The packing material utilized by Shields et al. was composed of oyster shells. A nutrient mineral solution containing yeast extract, peptone, glucose, and lactic acid was continuously recirculated through the

column. After a 48 hour inoculation period, 90% TCE removal was demonstrated over four days at an inlet TCE concentration of 130 μg/L (25 ppmv) and a 4-h vapor contact time (Shields et al., 1993). Govind et al. (1993) demonstrated sustained TCE removal from a mixed wastestream using a biotrickling filter containing activated carbon pellets as the packing material. Toluene within the vapor stream was utilized as the cosubstrate at a concentration of 520 ppmv. Govind et al. (1993) demonstrated sustained TCE and toluene removal efficiencies of greater than 95% for over 3 months at vapor contact times between 2 and 4 min and an inlet TCE concentration of 25 ppmv. However, when a ceramic packing material was used, only 30 to 40% TCE removal was observed (Govind et al., 1993). This difference in performance was attributed to adsorption of the cosubstrate (toluene) onto the carbon packing, which allowed for a low level of toluene usage by the TCE-degrading microorganisms in sections of the column where very little toluene remained in the vapor. In the system containing the ceramic packing, toluene was removed almost completely at the entrance of the column, and little toluene was available in the other portions of the column for TCE removal. More recently, Govind et al. (1995) have demonstrated removal of TCE and PCE within a biotrickling filter using microorganisms encapsulated within a hydrogel. The removal of TCE and PCE was attributed to partial reductive dechlorination caused by anaerobic conditions within the interior of the hydrogel created by consumption of oxygen at the surface. Greater than 99% TCE removal was demonstrated at a vapor contact time of 1.2 min for an inlet TCE concentration of 25 ppmv and an inlet toluene concentration up to 100 ppmv. Greater than 99% PCE removal was demonstrated at a vapor contact time of 4 min for an inlet PCE concentration of 25 ppmv and an inlet toluene concentration up to 100 ppmv (Govind et al., 1995).

C. BIOSCRUBBERS

A third type of biological system used for treating contaminated vapor streams is the bioscrubber. Bioscrubbing is a process whereby absorption of vapor-phase contaminants into an aqueous phase occurs, with the subsequent oxidation of the contaminants by microorganisms suspended in the liquid (Dharmavaram, 1991; van Groenestijn and Hesselink, 1993; VDI Richtlinie 3478, 1985).

For most bioscrubbing systems, absorption and biological oxidation are separated into two distinct unit operations, with absorption of the contaminants into water occurring in a scrubber column, followed by the degradation of the contaminant in a biological reactor (Dharmavaram, 1991; van Groenestijn and Hesselink, 1993; VDI Richtlinie 3478, 1985; Overcamp et al., 1993). The biological reactor is frequently an activated sludge system (VDI Richtlinie 3478). Using mathematical modeling, Overcamp et al. (1992) have found that bioscrubbers are very economical for degrading highly soluble

contaminants, such as alcohols, whereas biotrickling filters should be used for treating less-water-soluble contaminants. However, the use of activated carbon in the recirculating water has recently been proposed as a way of enhancing the solubility and degradability of sparingly soluble contaminants (Overcamp et al., 1994; Kok, 1994). Bioscrubbers typically have been used for treating odors, but a recent bioscrubbing application includes treating ethanol emissions from a brewery (Croonenberghs et al., 1994).

Envirogen also has demonstrated that bioscrubbers can be designed so that absorption into water and biological oxidation occur concurrently in the same reactor. By designing the system in this manner, the consumption of the contaminant in the liquid droplets by the microorganisms can be used to increase the mass transfer rate into the droplets. In addition, mass transfer appears to be enhanced by small liquid droplets due to adsorption effects. It has been observed that "fog," or small aqueous droplets 10 μm or less in diameter, absorb vapor contaminants with much higher efficiency, sometimes two or four orders of magnitude higher, than would be predicted based on Henry's law alone (Capel et al., 1991; Glotfeldy et al., 1990; Glotfeldy et al., 1987).

D. SUSPENDED-GROWTH REACTORS

A fourth type of biological system used for treatment of contaminated vapor streams is the suspended-growth reactor (Dasu et al., 1993; Ensley and Kurisko, 1994). These reactors may be bubble columns or stirred tanks, and may even contain activated carbon as a biomass support matrix (Ye et al., 1994). These systems are most effective when a very high degree of process control is required, and are most economical for treatment of low-volumetric-vapor flowrates (100 to 200 cfm) containing high concentrations of contaminants (greater than 1,000 ppmv as C) (Radian/Envirogen, 1996; Envirogen, 1996).

Greater than 90% TCE removal has been demonstrated using the bubble column design when treating a TCE/benzene stream (Folsom, 1992), when treating a wastestream containing other chlorinated solvents (DOE, 1994), and when treating a pure TCE wastestream at concentrations 5 to10 mg/L of TCE in the vapor phase (Ensley, 1992). Effective long-term performance for over 9 months, averaging over 90% TCE removal, was demonstrated with the stirred-tank design (Envirogen, 1996). In these studies, *P. cepacia* strain G4 and/or *P. mendocina* were used along with phenol (for *P. cepacia* G4) and/or toluene (for both *P. cepacia* and *P. mendocina*) as cosubstrates for TCE degradation. The stirred-tank reactor design and TCE treatment process have been effectively demonstrated in the field at Robins AFB, Georgia, and at F.E. Warren AFB, Wyoming using a 1,000-gallon tank (Envirogen, 1996;

Radian/Envirogen, 1996). At F.E. Warren AFB, 85 to 90% total TCE removal was demonstrated over a 70-day period (Radian/Envirogen, 1996).

E. MEMBRANE BIOREACTORS

Membrane biofilters employ semipermeable membranes that are impermeable to water. The biological aqueous phase is on one side of the biofilm while the gas phase is on the other side. Organic contaminants and oxygen diffuse through the membrane, from the gas phase to the aqueous phase, where they are degraded. Currently, membrane bioreactors are also being investigated for treatment of contaminated vapors (McGrath and Ergas, 1995; Reij et al., 1995a). These systems operate as fixed-film systems, with biomass growing on the "tube" or "shell" side of a membrane-contacting device. As a vapor stream flows through either the "shell" or "tube" side of the system, mass transfer of the contaminants across the membrane boundary occurs, with the subsequent degradation of the contaminants in the biofilm. The biofilm is kept moistened by a flow of recirculating medium across the biofilm layer. The medium is also used for pH and other process control. This technology is relatively new, and the range of applicability has not yet been determined.

SECTION V
BIOGAS REACTOR TECHNOLOGY

Specific process control parameters will influence the biological reactor process selection and its design. Those parameters include contaminant mass transfer requirements from the gas phase to the aqueous phase, cosubstrate feeding and process control requirements, pH and alkalinity control, target organic contaminant DREs, and volumetric flow capacity. An additional consideration is the potential for anaerobic biological activity to promote CAC dechlorination in the reactors. The four reactor configurations being considered for the off-gas treatment process are conventional biofilters, fixed-film trickling filters, bioscrubbers, and complete-mix reactors with air sparging. Membrane bioreactors also offer attractive advantages for off-gas biological treatment, but they remain in their infancy technologically and would require significantly more development before being considered for such a complex off-gas stream.

As discussed in the previous sections, the McClellan AFB off-gas contains a very complex mixture of contaminants, including some CACs that are relatively difficult to degrade. However, many of these CACs are degraded cometabolically by bacteria that grow on methane, ammonia, propane, propylene, toluene, phenol, or IPB. One major contaminant, PCE, can be degraded only under anaerobic conditions via reductive dechlorination. The system configuration must be capable of (1) allowing for the effective addition of the cosubstrate and its efficient degradation within the biological reactor; (2) controlling process pH, alkalinity, temperature, and other physical/chemical parameters; and (3) economically processing the required vapor throughput. Another design consideration is the possible development of anaerobic zones within a biofilm to promote reductive dechlorination.

A. CONTAMINANT MASS TRANSFER REQUIREMENTS

The biological treatment of the SVE off-gas and the efficient removal of the contaminants from the gas phase require the efficient mass transfer of the organic contaminants to the aqueous phase of the reactor. Bioscrubbers rely on the mass transfer of the contaminants from the off-gas to the aqueous phase, followed by the biological treatment of the contaminated effluent water in a separate stage. The efficient use of an aqueous bioscrubber requires that the contaminants have relatively low Henry's coefficients of less than 10^{-3} atm-m^3/mol. Removal of compounds that have Henry's constants greater than 10^{-3} atm-m^3/mol is limited by mass transfer, resulting in low removal efficiencies ($\leq$ 50%) (Overcamp et al., 1993). Except for acetone and the ketones, most of the compounds present in the

McClellan AFB off-gas stream are sparingly soluble in water and have Henry's constants greater than 10^{-3} atm-m^3/mol. A bioscrubber would require in excess of 3,500 gpm of water to strip the contaminants from the off-gas (at a flow rate of 1000 scfm), indicating that a bioscrubber would not be an effective system for the removal of the organic contaminants from the off-gas. Thus, a bioscrubber system is not recommended for the off-gas treatment at McClellan AFB.

Conventional biofilters, complete-mix suspended-growth bioreactors, and biotrickling filters also rely on the efficient mass transfer of contaminants from the gas phase to the aqueous phase. Bioscrubbers rely on the separate removal of the contaminants into the aqueous phase and subsequent treatment of the contaminated water. The adsorption onto organic solids and biological activity contribute to the maintenance of very low aqueous-phase organic contaminant concentrations, and the low aqueous-phase concentrations produce a large concentration gradient between the gas and aqueous phases, resulting in more efficient mass transfer.

TCE degradation has been demonstrated using complete-mix, suspended-growth bioreactors (Ensley, 1992) and biotrickling filters (Govind et al., 1993; Govind et al., 1995) with DREs greater than 90%. Therefore, both systems are considered further for the McClellan AFB application.

B. REQUIREMENTS FOR COSUBSTRATE FEEDING AND CAC DEGRADATION RATES

Because a significant fraction of the McClellan AFB SVE off-gas contaminants must be treated cometabolically, cosubstrate feeding is an important consideration in the system configuration. The low concentrations of toluene (and possibly other aromatic hydrocarbon compounds) already present in the wastestream may contribute to a small fraction of the overall CAC cometabolic degradation, but the addition of another cosubstrate will be required for complete CAC degradation.

Both gaseous (methane, propane, and/or ammonia) and liquid (phenol, toluene, and/or IPB) cosubstrates can be readily added to the process water of complete-mix bioreactors and biotrickling filters.

The ability to control the cosubstrate feeding rate and the steady-state reactor cosubstrate concentration is vital to the efficient operation of the biological treatment process and the ability to achieve high DREs. The steady-state cosubstrate concentration within the reactor must be kept low to prevent competitive inhibition between the cosubstrate and the CACs, whereas sufficient cosubstrate must be added to the process to sustain an active biomass for cometabolic CAC degradation. Thus, careful process control of the cosubstrate fed to the reactor system is necessary. Steady-state cosubstrate concentrations must be controlled by influent flowrates and the liquid recycle rate. Liquid cosubstrates

can be metered into the process water of biotrickling filters and suspended-growth bioreactors with relative ease; liquid cosubstrates cannot be added to biofilters easily, because biofilters are designed to operate with a stationary water phase and do not employ a liquid recycle system.

Introducing methane and propane into the process water is more difficult than adding liquid cosubstrates because methane and propane have high Henry's constants, and they do not readily partition from the gas phase into the aqueous phase. Thus, they cannot simply be metered into the influent gas stream because they will not partition into the aqueous phase quickly enough and will not be made available to the bacteria. Thus, a substantial fraction of the mass of cosubstrate added to the system will pass through the reactors unused.

A more effective method of adding gases to process water is to use a high-efficiency mass-transfer bubble contactor, similar to contactors used to supply pure oxygen to water in aerobic reactor systems (Sutton and Mishra, 1994). Oxygen bubble contactors can be incorporated for methane or propane addition to bioreactors.

As mentioned earlier, low cosubstrate concentrations must be maintained to minimize competitive inhibition effects on CAC degradation. Complete-mix bioreactors provide a well-mixed system that results in rapid dilution of influent substrates and low effluent concentrations, but they require relatively rapid substrate degradation rates. Complete-mix reactors are less effective for compounds that exhibit first-order degradation characteristics, such as TCE (and presumably other CACs). Plug-flow reactors, on the other hand, are much more effective for compounds with first-order degradation rates. Biotrickling filters offer a plug-flow configuration. One disadvantage of plug-flow reactors is that the substrate concentrations entering a reactor can be too high. High influent concentrations could contribute to competition between cosubstrate and CACs. By increasing the liquid recycle rate, the liquid phase of the plug-flow reactor can be mixed to dilute the influent cosubstrate concentration. At the same time, the gas phase can be operated under plug-flow conditions.

C. ALKALINITY REQUIREMENTS AND pH CONTROL

Conventional compost-based biofilters typically are not used for treatment of chlorinated compounds due to their inability to effectively control pH. Biotrickling filters and suspended-growth reactors are capable of effectively controlling pH by adding alkalinity (bicarbonate or carbonate) to the process water or by titrating NaOH into the process water using an electronic feedback system that monitors pH. The NaOH titration involves more risk than the buffering system, but is more economical

for biological processes treating wastes with high organic chlorine concentrations and significant acid production.

D. FLOW CAPACITY

Envirogen has modeled TCE degradation within its stirred-tank configuration using phenol as the cosubstrate and has shown that TCE removal is not limited by mass transfer, but is primarily limited by the biological degradation rate. Assuming a volatile suspended solids (VSS) concentration of 5,000 mg/L, 500 ppmv TCE in the gas phase, and 90% TCE removal, the maximum volume of air that that can be processed through a complete-mix bioreactor is 0.7 to 0.14 volume of contaminated air per volume of liquid suspension per minute (vol/vol-min) (Envirogen, 1996; Radian/Envirogen, 1996). These flowrates correspond to an effective vapor contact time of 7 to 14 minutes (i.e., reactor volume divided by the vapor flowrate). For 95% TCE removal, the effective vapor contact time increases to 50 min (Envirogen, 1996). Thus, a 600-scfm reactor would require approximately 4,300 to 8,500 ft^3 to achieve 90% CAC removal, and up to 7 times those volumes to achieve 95% removal.

In contrast to the volume required for the complete-mix system, Govind et al. (1993) demonstrated 95% TCE removal using a biotrickling filter with an inlet TCE concentration of 25 ppmv. The reactor was operated with a 2- to 4-min vapor contact time (Govind et al., 1993). The lower contact times demonstrated for the biotrickling filter are attributed to the first-order TCE degradation kinetics; as mentioned above, a plug-flow reactor is expected to outperform a complete-mix reactor when reaction rates are first order, given similar vapor contact times (Andrews and Noah, 1995). In addition, because biotrickling filters employ a fixed-film process, the effective biomass concentration per unit volume of reactor can be maintained much higher than for a suspended-growth reactor (Andrews and Noah, 1995), resulting in higher performance per unit reactor volume.

E. CAPACITY FOR ANAEROBIC TREATMENT ZONES

As discussed in Section III, some of the compounds in the SVE off-gas stream can be dechlorinated biologically under anaerobic conditions. Two compounds, PCE and Freon™ 113, are aerobically recalcitrant and can be biotransformed only anaerobically. Although the biological processes will be operated aerobically, there is a strong possibility that anaerobic regions will be formed in a biological trickling filter process, whereas no such regions are possible in a complete-mix system. In the deeper layers of a biofilm, anaerobic areas where sulfate-reducing bacteria and methanogens can survive

can be formed due to oxygen mass-transfer limitations in the biofilm (Arvin and Harremoes, 1990; Baltzis and Shareefdeen, 1994; Enzien et al., 1994). Enzien et al. (1994) reported the biodegradation of TCE and PCE under aerobic conditions in a sediment column; *c*-DCE was the major product of TCE and PCE dechlorination, no VC was detected, and the bulk liquid DO concentration was greater than 1.6 mg/L. In spite of the bulk aerobic conditions, methane was detected in the pore waters. The authors attributed the dechlorination to the development of anaerobic conditions in "microsites" within the soil matrix. To date, anaerobic sites within biofilms have not been exploited to promote reductive dechlorination of CACs under bulk aerobic conditions. Anaerobic pockets will not be a controlling process selection and design factor for the off-gas biological treatment system, but the bioreactor design may include the flexibility of enhancing anaerobic activity via process control during the laboratory-phase study.

SECTION VI
TECHNOLOGY SELECTION AND LABORATORY EVALUATION

Based on the discussion in Section V, the biotrickling filter is the most suitable reactor configuration for the McClellan AFB SVE off-gas application. Biotrickling filters (1) can be operated with high contaminant mass-transfer rates, (2) allow for the efficient addition and control of liquid- or gas-phase cosubstrates, (3) provide a plug-flow configuration for efficient degradation of contaminants that exhibit first-order degradation rates, (4) permit relatively simple pH control via alkalinity addition or NaOH titration, (5) economically process the required vapor throughput with low vapor-contact times, and (6) provide for the possibility of reductive dechlorination via the production of anaerobic pockets within the biofilm. Two parallel trickling biofilters will be operated in the laboratory. The two biotrickling filter units will be used to test different cosubstrates and different operating parameters to optimize vapor throughput, contaminant DREs, and cosubstrate use. In this section, the biotrickling filter design approach is presented in greater detail.

A. COSUBSTRATE SELECTION

Cosubstrate selection is an important design consideration for the off-gas wastestream. The cosubstrate-degrading bacteria must be able to degrade chloroethenes and chloroethanes, they must be able to degrade CACs and the cosubstrate simultaneously, and they must be resistant to CAC intermediate toxicity. Among the cosubstrates reported in the literature including methane, propane, propylene, ammonia, toluene, phenol, and IPB, only methanotrophs, propane-oxidizers, ammonia-oxidizers, and IPB-oxidizers have been shown to degrade chloroethenes and chloroethanes. Ammonia is not suitable due to severe competitive inhibition expressed by ammonia on TCE degradation rates. Methane is unsuitable because methanotrophic cultures are relatively susceptible to intermediate product toxicity, exhaustion of intracellular energy (i.e., NADH), and inhibition of sMMO production in the presence of high copper concentrations.

IPB and propane remain as potential cosubstrates. The fact that limited information is available on both cosubstrates is a concern, but does not outweigh the fact that both cosubstrates support chloroethene and chloroethane degradation, and both have been demonstrated to resist intermediate toxicity. IPB has the added advantage of being soluble, simplifying its addition to the liquid stream.

Because of the importance of the cosubstrate and the development of a mixed biological culture capable of efficient CAC degradation, flexibility must be incorporated into the system design to be able

to change cosubstrates during the study should either propane or IPB fail to meet the DRE requirements. If one or both cosubstrates fail to promote CAC degradation, another cosubstrate will be selected and reactor operating parameters will be revisited at that time. The most likely alternative cosubstrate is methane. Toluene and phenol cannot be used because they do not support the degradation of chlorinated ethanes. Ammonia is not likely to be considered because ammonia oxidizers are relatively sensitive to CAC intermediate toxicity, and there is competition between ammonia and CACs. In addition to the cosubstrate selection, the cosubstrate/contaminant feeding ratio (R_s) is also an important design consideration. This ratio is the inverse of the transformation yield (T_y), used by Dolan and McCarty (1995). Using propane as a cosubstrate, Phelps et al. (1990) reported a value of 26 to 34 g of propane consumed per g of TCE degraded, whereas Chang and Alvarez-Cohen (1995) reported a value of 43 g of propane per g of TCE degraded.

R_s values for IPB have not been reported in the literature, but their values are expected to be similar to those of phenol and toluene, because all three are aromatic compounds and are structurally similar. For toluene, Chang and Alvarez-Cohen (1995) reported a value of 21 g of toluene consumed per g TCE degraded for a mixed toluene-degrading culture; Ensley (1992) reported an average value of 22 g of toluene per g TCE degraded for *P. mendocina* in a bubble-column reactor; Govind et al. (1993) reported 15 g of toluene per g of TCE in biotrickling filters. For phenol, Hopkins et al. (1993b) reported a value of 16 g of phenol per g of TCE during a field-pilot test, and 9 g of phenol per g TCE degraded for resting cells in the laboratory. Ensley (1992) reported an average value of 6 g of phenol per g TCE degraded for *P. cepacia* G4 in a bubble column reactor and saw TCE degradation at phenol concentrations as low as 2 g phenol per g TCE.

The values described above do not necessarily represent optimal R_s values, because not all studies attempted to optimize the cosubstrate-to-TCE ratio. The initial cosubstrate to CAC ratios for propane and IPB during the laboratory-phase tests will be based on the values reported above for propane and phenol (i.e., 30 g propane per g CAC and 6 g IPB per g CAC), assuming IPB concentrations should be similar to phenol concentrations under similar operating conditions. The low R_s value for IPB is based on Envirogen's experience using phenol as the cosubstrate for TCE degradation within stirred-tank reactors. The more conservative value for propane will be used because there is limited background literature that describes the use of propane as a cosubstrate for cometabolism of CACs in biological reactors. In addition, the excess propane will be needed to maintain adequate propane concentrations in the aqueous phase of the reactor. Because of the scale of the laboratory reactors, a high-efficiency mass transfer device will not be employed for propane during the laboratory phase, and propane will be added

in the gas phase. This will result in less efficient propane utilization, but will be less expensive and easier to control in the laboratory.

Aqueous-phase propane concentrations will be controlled by regulating the partial pressure of propane in the gas phase. Aqueous-phase IPB concentrations will be controlled by regulating the IPB feed rate via the feed concentration, and by regulating the liquid recycle rate.

B. SYSTEM CONFIGURATION

Selective pressures in a complex, mixed biological community will result in the growth and proliferation of bacteria that receive the greatest benefit from the available substrate(s), followed by the growth of bacteria that gain progressively less energy for cell growth and maintenance. In the absence of a cosubstrate, the NCOCs, including acetone, BTEX, and ethyl toluene, will be degraded first by bacteria that can beneficially exploit energy and carbon for growth from those compounds. Bacteria that degrade MEK or MIBK will grow next, followed by those that degrade DCBs. This kind of stratification is based on the available energy from each group of compounds and the relative ease for the bacteria to exploit that energy.

Addition of the cosubstrate in the presence of the NCOCs and DCBs in the off-gas may result in biological competition between those compounds in the biotrickling-filter reactor. For example, if IPB is added before the acetone is degraded, bacteria that can degrade either IPB or acetone will most likely preferentially degrade the acetone, followed by IPB. On the other hand, those that degrade both IPB and DCBs may be expected to preferentially degrade IPB, leaving the DCBs undegraded until the IPB is exhausted from the liquid stream. Thus, the addition of the cosubstrate in the presence of other growth-related substrates may result in competition between those substrates and in the inefficient use of the cosubstrate. Because CACs do not provide carbon and energy for cell growth, their degradation is not expected to confer any selective advantage to the CAC-degrading bacteria. Therefore, the cosubstrate should be added to the biotrickling filter column after the gas-phase growth substrates (acetone, BTEX, ethyl toluene, MEK, MIBK, and DCBs) have been consumed. This will be accomplished by staging the biotrickling filter. Two-stage columns will be used. The first stage will not be fed a cosubstrate and will be used to promote the degradation of growth-related contaminants in the off-gas. The removal of those contaminants will result in a simplified gas stream that will be fed to the second stage. The second stage will be used to promote cometabolic CAC degradation, using either propane or IPB as cosubstrates. Figure 1 shows a schematic of the proposed process configuration. A more detailed diagram is provided in the attached Laboratory operation schedule. Independent liquid feed and recycle streams will be

maintained for each reactor stage. Alkalinity and pH also will be controlled independently for each stage. The gas will be fed to the bioreactors cocurrent with the direction of the liquid flow, which will be fed to the top of each reactor and allowed to trickle down via gravity flow. The off-gas from the first stage will be fed to the second stage. At a minimum, the reactors will be monitored at the influent and effluent of both reactor stages.

C. VAPOR THROUGHPUT

The vapor throughput of the biotrickling filter systems will be dictated by the vapor contact time required for contaminant removal to predetermined DREs (i.e., 95%). The vapor contact time is defined as the reactor volume occupied by packing divided by the vapor flowrate. The reactor dimensions (i.e., the height-to-diameter ratio) are dictated by the maximum allowable pressure drop and head loss for the reactor system. For TCE degradation, Govind et al. (1993) demonstrated 95% TCE removal in a biotrickling filter system containing activated carbon as the packing material and using an empty bed vapor contact time of 2 to 4 min. Because carbon, which can provide buffering capacity for shock loads, will not be used for this application, and because of the complexity of the wastestream, a longer vapor contact time of 4 to 5 min will be employed during the laboratory-phase test. Vapor throughput will be adjusted to maximize the DREs, as necessary. In addition, vapor throughput will be optimized during the process optimization period to maximize the throughput while maintaining target DREs (see the attached laboratory operations schedule in Appendix A).

D. LIQUID (AND BIOMASS) DISCHARGE

Liquid must be discharged from the biotrickling filter system, either continuously or intermittently, to prevent the accumulation of sodium chloride (NaCl) and the exhaustion of inorganic nutrients supplied in the process water. NaCl will be produced from chloride ions released during CAC degradation. The NaCl concentration will be maintained below 2%. However, NaCl concentrations in biofilters have been reported to reach as high as 2.2 to 3.5% (Diks et al., 1994a) and Environgen, Inc. has operated biofilters with NaCl concentrations as high as 2.5% without adversely affecting the contaminant degradation characteristics. The liquid discharge also will contain low biomass concentrations. The biomass concentration will depend on the cosubstrate mass fed to the reactors and the biomass yield of the cosubstrate.

Continuous liquid wasting also will prevent the accumulation of process water fed to the system. The system waste rate will be adjusted to prevent NaCl accumulation, to maintain low aqueous-phase cosubstrate concentrations, to allow sufficient cosubstrate and inorganic nutrient feeding, and to control biomass sloughing from the biofilm.

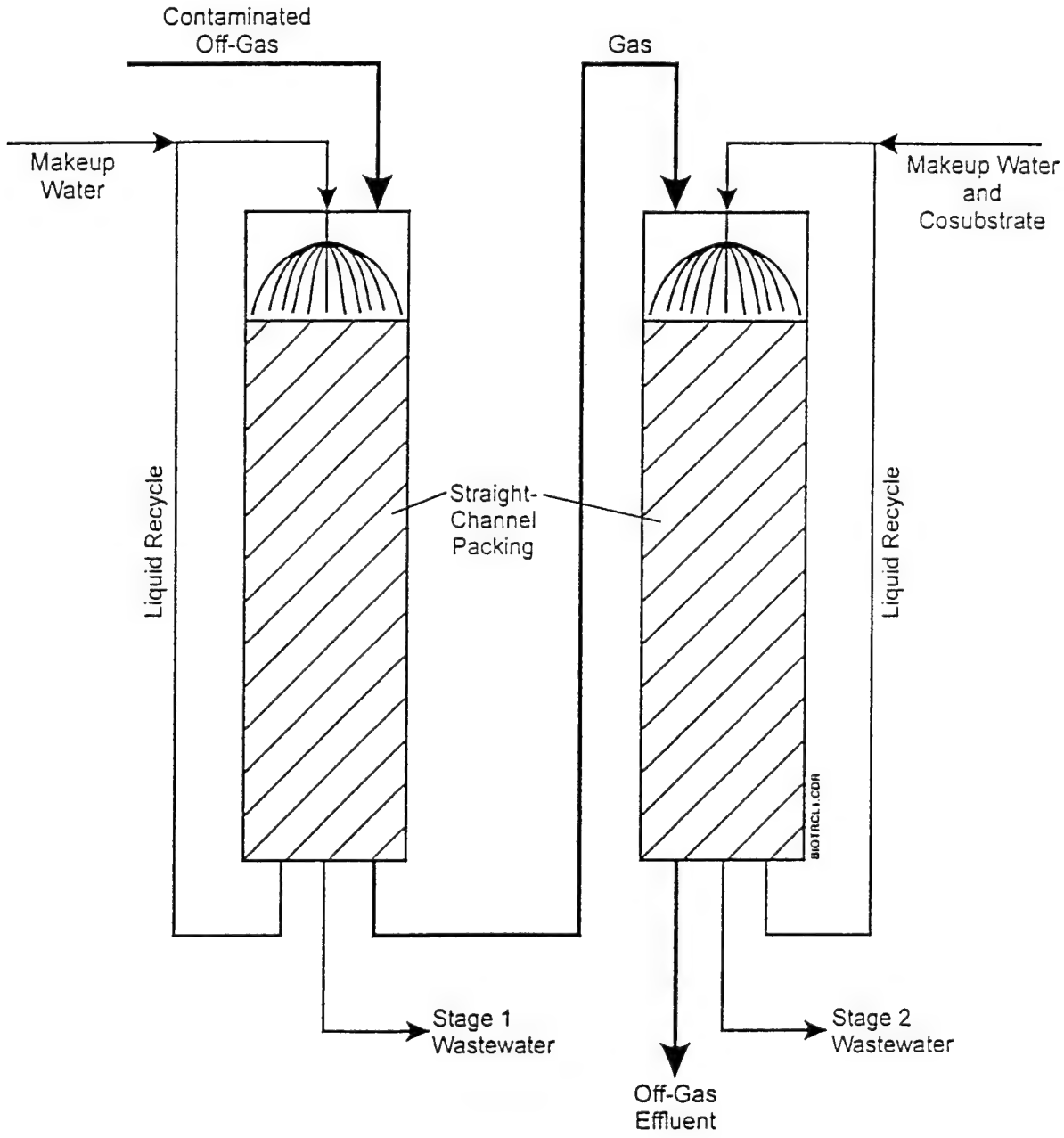

Figure 1. Biotrickling Filter Reactor System Configuration

E. REACTOR PLUGGING

The biotrickling filter systems will be designed to prevent biomass plugging and excessive packing pressure drops. This will be accomplished by operating the systems in a downflow configuration at appropriate liquid and vapor velocities. Biomass growth is controlled by the liquid and vapor shear forces. Because of scale-up limitations, the bench-scale systems will not be operated to control pressure drop. However, the pressure drop across the packing will be monitored in the laboratory to provide information that will contribute to the scale-up to pilot- and full-scale systems, along with packing material-specific pressure drop curves.

For the McClellan AFB application, biomass growth also may be controlled by the relatively high NaCl concentrations (Diks et al., 1994a). The actual benefit of the high NaCl concentrations for the control of the reactor biomass processes cannot be determined at this time, but NaCl concentrations will be monitored closely and adjusted for maximum control.

F. SYSTEM OPTIMIZATION

The system will be optimized to maximize vapor throughput, maximize DREs, minimize cosubstrate use, and promote anaerobic activity within the biofilm. Vapor throughput will be maximized by increasing vapor flowrates to determine the minimum vapor contact time required to achieve the desired DREs. Vapor throughput and DREs will be interdependent, and changing one will most likely change the other. DREs will be maximized by controlling vapor throughput for the two stages, maintaining optimal pH and NaCl concentrations, and adjusting cosubstrate addition as appropriate. Cosubstrate use will be optimized by determining the minimum cosubstrate loading rate while maintaining predetermined DREs by progressively decreasing cosubstrate usage at a fixed vapor throughput rate.

Anaerobic zones in the biofilm may be exploited for the degradation of PCE, TCE, 1,1,1-TCA, and/or Freon™ 113. The anaerobic zones may be created in the deeper layers of the biofilm in the biotrickling filters, creating areas where anaerobic bacteria can survive and proliferate (Arvin and Harremoes, 1990; Enzien et al., 1994). Reductive dehalogenation may occur in either the first or second stage. The first stage will be supplied with easily degraded organic substrates in the gas phase as potential electron donors for reductive dehalogenation, and the second stage will be supplied with an organic cosubstrate that also may act as an electron donor. In the absence of anaerobic activity during normal operation, an easily degraded growth substrate, such as lactate, may be added to the recirculating

water in either stage to promote a thicker biofilm and anaerobic activities within the biofilm. Anaerobic activity will be monitored by monitoring influent and effluent PCE.

SECTION VII
CONCLUSIONS

Based on the characteristics of the off-gas stream and a review of available biological treatment options, the biotrickling filter process configuration was selected as the best process for the McClellan AFB SVE off-gas stream. Conventional compost-based biofilters would require frequent bed replacement, and the high Henry's law constants of most of the contaminants in the off-gas prevent the economical use of a bioscrubber system. Slow, first-order degradation rates of the CACs in the off-gas prevented the selection of a complete-mix suspended-growth biological reactor.

Because of the complexity of the McClellan AFB off-gas stream, and because of presence of growth-supporting contaminants as well as contaminants that do not support growth, a two-stage biotrickling filter system is proposed. The first stage will be used to remove the easily degraded growth substrates from the stream, such as acetone, BTEX, MEK, MIBK, and DCBs. The second stage will be used to cometabolically degrade CACs in the off-gas. Two cosubstrates will be investigated initially, using two separate two-stage reactor units. Propane and IPB were selected as the cosubstrates for this investigation because of their abilities to support bacteria that can degrade chloroethenes and chloroethanes, and because of their potential resistance to intermediate toxicity and competitive inhibition with CACs. Propane will be added to the influent gas stream, and IPB will be metered into the liquid recirculation line.

The two separate reactor units will provide greater opportunity to optimize the system with respect to vapor throughput, contaminant DREs, cosubstrate use, and possibly anaerobic activity to promote reductive dechlorination in the anaerobic zones within the biofilm. As a starting point for bench-scale optimization, a vapor contact time of 4 to 5 min will be employed. Initially, a propane-to-CAC ratio of 30 will be used, and an IPB to CAC ratio of 6 will be used. Aqueous-phase propane concentrations will be controlled by adjusting the partial pressure of propane in the gas phase. Aqueous-phase IPB concentrations will be controlled by controlling the influent IPB loading rate and the liquid recirculation rate. The contaminated off-gas stream will be fed into the system with a single pass; the gas phase will not be recirculated in the system.

The liquid removal rates from each stage will be set initially to maintain an NaCl concentration below 2.0%. Optimization may include the use of powdered activated carbon (PAC) in the recirculation water in the second stage of both systems to investigate the effects of PAC on system performance. PAC may enhance the solubility and degradability of sparingly soluble contaminants and can protect the

system against organic shock loads. It also can provide a method of regulating the supply of cosubstrate to the reactors by having a cosubstrate/PAC contact unit. Another unique treatment approach is the use of high-substrate feed zones to stimulate anaerobic activity in the biofilm for anaerobic reductive CAC dechlorination.

SECTION VIII

REFERENCES

Allen, E.R., and Y. Yang. 1992. "Biofiltration: An Air Pollution Control Technology for Hydrogen Sulfide Emissions," in *Industrial Environmental Chemistry*, D.T. Sawyer and A.E. Martell, Eds., Plenum Press, New York, pp. 273-287.

Alvarez-Cohen, L., and P.L. McCarty. 1991a. "Effects of Toxicity, Aeration, and Reductant Supply on Trichloroethylene Transformation by a Mixed Methanotrophic Culture." *Appl. Environ. Microbiol. 57*(1):228-235.

Alvarez-Cohen, L., and P.L. McCarty. 1991b. "Two-Stage Dispersed Growth Treatment of Halogenated Aliphatic Compounds by Cometabolism." *Environ. Sci. Technol. 25*:1387-1393.

Anderson, W.C. 1994. *Innovative Site Remediation Technology*. American Academy of Environmental Engineers. Annapolis, MD.

Andrews, G.F., and K.S. Noah. 1995. "Design of Gas-Treatment Bioreactors." *Biotechnol. Prog. 11*(5):498-509.

Apel, W.A., W.D. Kant, F.S. Colwell, B. Singleton, B.D. Lee, G.F. Andrews, A.M. Espinosa, and E.G. Johnson. 1993. *Removal of Gasoline Vapors from Air Streams by Biofiltration*. Report No. EGG-WTD-10714, Idaho National Engineering Laboratory, EG&G Idaho, Inc.

Arciero, D., T. Vannelli, M. Logan, and A.B. Hooper. 1989. "Degradation of Trichloroethylene by the Ammonia-Oxidizing Bacterium *Nitrosomonas Europaea*." *Biochemical and Biophysical Research Communications. 159*(2):640-643.

Arvin, E., and P. Harremoes. 1990. "Concepts and Models for Biofilm Reactor Performance." *Wat. Sci. Tech. 22*(1-2):171-192.

Baltzis, B.C., and Z. Shareefdeen. 1994. "Biofiltration of VOC Mixtures: Modeling and Pilot Scale Experimental Verification." Paper no. 94-TA260.10P, 87th Annual Air & Waste Management Association Meeting & Exhibition, Cincinnati, OH, June 19-24.

Begley, R. 1991. "Industry Watches Closely as EPA Converts the Clean Air Act into Regulation." *Chemical Week 13*:23-28.

Bielefeldt, A.R., H.D. Stensel, and S.E. Strand. 1995. "Cometabolic Degradation of TCE and DCE Without Intermediate Toxicity." *J. of Environ. Engin. 121*:791-797.

Bohn, H. 1992. "Consider Biofiltration for Decontaminating Gases." *Chem. Eng. Prog. 88*(4):34-40.

Bohn, H., and R. Bohn. 1988. "Soil Beds Weed Out Air Pollutants." *Chemical Engineering 95*(4):74-76.

Broholm, K., T.H. Christensen, and B.K. Jensen. 1993. "Different Abilities of Eight Mixed Cultures of Methane-Oxidizing Bacteria to Degrade TCE." *Wat. Res. 27*(2):215-224.

Capel, P., Leunberger, and Giger. 1991. "Hydrophobic Organic Chemicals in Urban Fog." *Atmospheric Environ. 25A*(7):1335-1346.

Castro, C.E., and N.O. Belser. 1990. "Biodehalogenation: Oxidative and Reductive Metabolism of 1,1,2,-Trichloroethane by *Pseudomonas Putida, Bioregeneration of Vinyl Chloride." Environmental Toxicology and Chemistry 9*:707-714.

Chang, H.L., and L. Alvarez-Cohen. 1995. "Model For The Cometabolic Biodegradation of Chlorinated Organics." *Environ. Sci. Technol. 29*:2357-2367.

Chen, C., J.A. Puhakka, and J.F. Ferguson. 1996. "Transformations of 1,1,2,2-Tetrachloroethane under Methanogenic Conditions." *Environ. Sci. Technol. 30*:542-547.

Chetty, A.S., J.A. Dyer, and K.L. Mulholland. 1992. "The Role of Economics in Selecting VOC Controls." Paper no. 92-114.13, 85th Annual Air & Waste Management Association Meeting & Exhibition, Kansas City, MO, June 21-26.

Croonenberghs, J., F. Varani, and P. Le Fevre. 1994. "Use of Bioscrubbing to Control Ethanol Emissions." Paper no. 94-RP115B.06, 87th Annual Air & Waste Management Association Meeting & Exhibition, Cincinnati, OH, June 19-24, 1994.

Dabrock, B., J. Riedel, J. Bertram, and G. Gottschalk. 1992. "IPB (cumene)—A New Substrate for the Isolation of Trichloroethene-Degrading Bacteria." *Arch Microbiol. 158*:9-13.

Dasu, B.N., V. Deshmane, R. Shanmugasundram, C.M. Lee, and K.L. Sublette. 1993. "Microbial Reduction of Sulfur Oxide and Nitric Oxide." *Fuel 72*:1705-1714.

de Bont, J.A.M., M.J.A.W. Vorage, S. Hartmans, and W.J.J. van den Tweel. 1986. "Microbial Degradation of 1,3-Dichlorobenzene." *Appl. Environ. Microbiol. 52*:677-680.

Debus, O., H. Baumgartl, and I. Sekoulov. 1994. "Influence of Fluid Velocities on the Degradation of Volatile Aromatic Compounds in Membrane Bound Biofilms." *Wat. Sci. Tech. 29*:253-262.

DeFilippi, L.J., M.B. Koch, C.M. Voellinger, D.R. Winstead, and F.S. Lupton. 1993. "A Biological Air Treatment System Based Upon the Use of a Structured Carbon Biomass Support." Presented at the IGT Symposium on Gas, Oil, and Environmental Biotechnology, Colorado Springs, CO, Nov. 29 - Dec. 1.

Devinny, J.S., and D.S. Hodge. 1995. "Formation of Acidic and Toxic Intermediates in Overloaded Ethanol Biofilters." *J. Air Waste Manage. Assoc. 45*(2):125-131.

Dharmavaram, S. 1991. "Biofiltration - A Lean Emissions Abatement Technology." Paper no. 91-103.2, 84th Annual Air & Waste Management Association Meeting & Exhibition, Vancouver, British Columbia, June 16-21.

Dharmavaram, S., U.C. Duursma, G. Rietbroek, and E. Waalewijn. 1995. "Use of a Biotrickling Filter (BTF) for Control of N,N-Dimethylacetamide Emissions." Paper no. 95-TA9B.01, 88th Annual Air & Waste Management Association Meeting & Exhibition, San Antonio, TX, June 18-23.

Diks, R.M.M., and S.P.P. Ottengraf. 1991a. "Verification Studies of a Simplified Model for the Removal of Dichloromethane from Waste Gases Using a Biological Trickling Filter (Part I)." *Bioprocess Engr. 6:*93-99.

Diks, R.M.M., and S.P.P. Ottengraf. 1991b. "Verification Studies of a Simplified Model for the Removal of Dichloromethane from Waste Gases Using a Biological Trickling Filter (Part II)." *Bioprocess Engr.* 6:131-140.

Diks, R.M.M., S.P.P. Ottengraf, and A.H.C. van den Oever. 1994a. "The Influence of NaCl on the Degradation Rate of Dichloromethane by *Hyphomicrobium* sp." *Biodegradation 5:*129-141.

Diks, R.M.M., S.P.P. Ottengraf, and S. Vrijland. 1994b. "The Existence of a Biological Equilibrium in a Trickling Filter For Waste Gas Purification." *Biotechnol. Bioeng. 44*:1279-1287.

Dolan, M.E. and P.L. McCarty. 1995. "Methanotrophic Chloroethene Transformation Capacities and 1,1-Dichlorethene Transformation Product Toxicity." *Environ. Sci. Technol. 29*:2741-2747.

DOE, see U.S. Department of Energy.

Dyer, J., and K. Mulholland. 1994. "Toxic Air Emissions: What is the Full Cost to Your Business?" *Environ. Engin. (supplement to Chemical Engineering)*, February, pp.4-8.

Ensign, S.A., M.R. Hyman, and D.J. Arp. 1992. "Cometabolic Degradation of Chlorinated Alkenes by Alkene Monooxygenase in a Propylene-Grown *Xanthobacter* Strain." *Appl. Environ. Microbiol. 58*(9):3038-3046.

Ensley, B.D. 1992. *Biodegradation of Chlorinated Hydrocarbons in a Vapor Phase Reactor*. DOE/CH-9207, prepared for the U.S. Department of Energy.

Ensley, B.D., and P.R. Kurisko. 1994. "A Gas Lift Bioreactor for Removal of Contaminants from the Vapor Phase." *Appl. Environ. Microbiol. 60*(1):285-290.

Envirogen. 1996. Final report for DoD contract FO8635-91-C-0198 [Phase II], "Liquid-phase Bioreactor for Degradation of Trichloroethylene and Benzene." In press.

Enzien, M.V., F. Picardal, T.C. Hazen, R.G. Arnold, and C.B. Fliermans. 1994. "Reductive Dechlorination of Trichloroethylene and Tetrachloroethylene Under Aerobic Conditions in a Sediment Column." *Appl. Environ. Microbiol. 60*(6):2200-2204.

Ergas, S.J., E.D. Schroeder, and D.P.Y. Chang. 1992. *Biodegradation Technology for Volatile Organic Compound Removal from Airstreams - Phase I: Performance Verification*." Final report for contract no. AO32-127. Prepared for the California EPA Air Resources Board, Research Division. May.

Ergas, S.J., E.D. Schroeder, and D.P.Y. Chang. 1993. "Control of Air Emissions of Dichloromethane, Trichloroethene, and Toluene by Biofiltration." Paper no. 93-WA-52B.01, 86th Annual Air & Waste Management Association Meeting & Exhibition, Denver, CO, June 13-18.

Ergas, S.J., K. Kinney, M.E. Fuller, and K.M. Scow. 1995. "Characterization of a Compost Biofiltration System Degrading Dichloromethane." *Biotechnol. Bioeng. 44*:1048-1054.

Fogel, M.M., A.R. Taddeo, and S. Fogel. 1986. "Biodegradation of Chlorinated Ethenes by a Methane-Utilizing Mixed Culture." *Appl. Environ. Microbiol. 51*(4):720-724.

Folsom, B. 1992. *Liquid-Phase Bioreactor for Degradation of Trichloroethylene and Benzene*. DoD final report no. AD-D-275035/4/XAB.

Folsom, B.R., and P.J. Chapman. 1991. "Performance Characterization of a Model Bioreactor for the Biodegradation of Trichloroethylene by *Pseudomonas cepacia* G4." *Appl. Environ. Microbiol. 57*(6):1602-1608.

Folsom, B.R., P.J. Chapman, and P.H. Pritchard. 1990. "Phenol and Trichloroethylene Degradation by *Pseudomonas cepacia* G4: Kinetics and Interactions Between Substrates." *Appl. Environ. Microbiol. 56*(5):1279-1285.

Fouhy, K. 1992. "Cleaning Waste Gas, Naturally." *Chemical Engineering 99*(12):41-46.

Fox, B.G., J.G. Borneman, L.P. Wackett, and J.D. Lipscomb. 1990. "Haloalkene Oxidation by the Soluble Methane Monooxygenase from *Methylosinus trichosporium* OB3b: Mechanistic and Environmental Implications." *Biochemistry 29*:6419-6427.

Freedman, D.L., and S.D. Herz. 1996. "Use of Ethylene and Ethane as Primary Substrates for Aerobic Cometabolism of Vinyl Chloride." *Water Environ. Res. 68*(3):320-328.

Galli, R., and P.L. McCarty. 1989. "Biotransformation of 1,1,1-Trichloroethane, Trichloromethane, and Tetrachloromethane by a *Clostridium* sp." *Appl. Environ. Microbiol. 55*(4):837-844.

Glotfeldy, D.E., M.S. Majewski, and J.M. Seiber. 1990. "Distribution of Several Organophosphorus Insecticides and Their Oxygen Analogues in a Foggy Atmosphere." *Environ. Sci. Technol. 24*(3):353-357.

Glotfelty, D.E., J.N. Seiber, and L.A. Lilijedahl. 1987. "Pesticides in Fog." *Nature 325*:602-605.

Govind, R., P.S.R.V. Prasad, and D.F. Bishop. 1995. "Anaerobic/Aerobic Degradation of Aliphatic Chlorinated Hydrocarbons in an Encapsulated Biomass Biofilter." Presented at the EPA Symposium on Bioremediation of Hazardous Wastes: Research, Development, and Field Evaluations, Rye Brook, NY, Aug. 8-10. Abstracts document no. EPA/600/R-95/076):50-52.

Govind, R., V. Utgikar, W. Zhao, Y. Shan, M. Parvatiyar, and D.F. Bishop. 1993. "Development of Novel Biofilters for Treatment of Volatile Organic Compounds (Organic contaminants)." Presented at the IGT Symposium on Gas, Oil & Environmental Biotechnology, Colorado Springs, CO, Nov. 29 - Dec. 3.

Griffiths, C., J. Graydon, M. de Menibus, and G. Boyer. 1995. "Methylotrophic Biofiltration of Chlorinated Aliphatic Hydrocarbons." Presented at the 1995 Battelle In-situ and On-site Bioreclamation Conference in San Diego, CA.

Haigler, B.E., S.F. Nishino, and J.C. Spain. 1988. "Degradation of 1,2-Dichlorobenene by a *Pseudomonas* sp." *Appl. Enviorn. Microbiol. 54*:294-301.

Harker, A.R., and Y. Kim. 1990. "Trichloroethylene Degradation by Two Independent Aromatic Degrading Pathways in *Alcaligenes eutrophus* JMP134." *Appl. Environ. Microbiol. 56*(4):1179-1181.

Hartmans, S., and J. Tramper. 1991. "Dichloromethane Removal from Waste Gases with a Trickle-Bed Bioreactor." *Bioprocess Eng. 6*:83-92.

Hartmans, S., and J.A.M. de Bont. 1992. "Aerobic Vinyl Chloride Metabolism in *Mycobacterium aurum* L1." *Appl. Environ. Microbiol. 58*(4):1220-1226.

HazTech News. 1996. "TVA Biofilter to be Tested (Cont.)." April 11, p. 52.

Heald, S., and R.O. Jenkins. 1994. "Trichloroethylene Removal and Oxidation Toxicity Medicated by Toluene Dioxygenase of *Pseudomonas putida*." *Appl. Environ. Microbiol. 60*(12):4634-4637.

Hecht, V., D. Brebbermann, P. Bremer, and W.D. Deckwer. 1995. "Cometabolic Degradation of Trichloroethylene in a Bubble Column Bioscrubber." *Biotechnol. Bioengin. 47*(4):461-469.

Heller, K. 1991. "Clean Air: A Fresh Challenge, Preempt the Rules with Volunteerism." *Chemical Week,* Nov. *13, pp.* 22-23.

Henry, S.M., and D. Grbi-Gali. 1991. "Influence of Endogenous and Exogenous Electron Donors and Trichloroethylene Oxidation Toxicity on Trichloroethylene Oxidation by Methanotrophic Cultures from a Groundwater Aquifer." *Appl. Environ. Microbiol. 57*(1):236-244.

Hodge, D.S., and J.S. Devinny. 1994. "Biofilter Treatment of Ethanol Vapors." *Environ. Prog. 13*(3):167-173.

Hodge, D.S., V.F. Medina, R.L. Islander, and J.S. Devinny. 1991. "Treatment of Hydrocarbon Fuel Vapors In Biofilters." *Environ. Technol. 12*:655-662.

Holubar, P., C. Andorfer, and R. Braun. 1995. "Prevention of Clogging in Trickling Filters for Purification of Hydrocarbon-Contaminated Waste Air." *Proceedings of the 1995 Conference on Biofiltration*, University of Southern California, October 5-6, pp. 115-121.

Hopkins, G.D., J. Munakata, L. Semprini, and P.L. McCarty. 1993a. "Trichloroethylene Concentration Effects on Pilot Field-Scale In-Situ Groundwater Bioremediation by Phenol-Oxidizing Microorganisms." *Environ. Sci. Technol. 27*:2542-2547.

Hopkins, G.D., L. Semprini, and P.L. McCarty. 1993b. "Microcosm and In Situ Field Studies of Enhanced Biotransformation of Trichloroethylene by Phenol-Utilizing Microorganisms." *Appl. Environ. Microbiol. 59*(7):2277-2285.

Hur, H.G., M.J. Sadowsky, and L.P. Wackett. 1994. "Metabolism of Chlorofluorocarbons and Polybrominated Compounds by *Pseudomonas putida* G786(pHG-2) via an Engineered Metabolic Pathway." *Appl. Environ. Microbiol. 60*(11):4148-4154.

Hyman, M.R., S.A. Russell, R.L. Ely, K.J. Williamson, and D.J. Arp. 1995. "Inhibition, Inactivation, and Recovery of Ammonia-Oxidizing Activity in Cometabolism of Trichloroethylene by *Nitrosomonas europaea.*" *Appl. Environ. Microbiol. 61*(4):1480-1487.

Janssen, D.B., A. Scheper, L. Dijkhuizen, and B. Witholt. 1985. "Degradation of Halogenated Aliphatic Compounds by *Xanthobacter authotrophicus* GJ10." *Appl. Environ. Microbiol. 49*(3):673-677.

Ju, L.-K., and G.G. Chase. 1992. "Improved Scale-Up Strategies of Bioreactors." *Bioprocess Engineering 8*:49-53.

Kampbell, D.H., J.T. Wilson, H.W. Read, and T.T. Stocksdale. 1987. "Removal of Volatile Aliphatic Hydrocarbons in a Soil Bioreactor." *JAPCA 37*(10):1236-1240.

Kok, H.J.G. 1994. "Bioscrubbing of Air Contaminated with High Concentrations of Hydrocarbons." *Proceedings of the International Symposium on Biological Waste Gas Cleaning*, Heidelberg, Germany, March 9-11. VDI Berichte No. 1104.

Kuter, G.A., J.E. Harper, L.M. Naylor, and P.J. Gormsen. 1993. "Design, Construction and Operation of Biofilters for Controlling Odors at Composting Facilities." Paper no. 93-WP-52C.02, 86th Annual Air & Waste Management Association Meeting & Exhibition, Denver, CO, June 13-18.

Lacky, L., and T. Holt. 1996. "Not for the Birds. Biofiltration Using Composted Chicken Litter Efficiently Removes Styrene from Industrial Exhaust Gases." *Industrial Wastewater,* May/June 1996, pp. 31-33.

Lam, T., and V.L. Vilker. 1987. "Biodehalogenation of Bromotrichloromethane and 1,2-Dibromo-3-chloropropane by *Pseudomonas putida* PpG-786." *Biotechnology and Bioengineering. 29:*151-159.

Landa, A.S., E.M. Sipkema, J. Weijma, A.A.C.M. Beenackers, J. Dolfing, and D.B. Janssen. 1994. "Cometabolic Degradation of Trichloroethylene by *Pseudomonas cepacia* G4 in a Chemostat With Toluene as the Primary Substrate." *Appl. Environ. Microbiol. 60*(9):3368-3374.

Langseth, S., and D. Pflum. 1994. "Weyerhaeuser Tests Large Pilot Biofilters for Organic Contaminants Removal." *Panel World,* March.

Lanzarone, N.A., and P.L. McCarty. 1990. "Column Studies on Methanotrophic Degradation of Trichloroethene and 1,2-Dichloroethane." *Ground Wat. 28*:810-891.

Leatherbarrow, R.J. 1990. "Use of Nonlinear Regression to Analyze Enzyme Kinetic Data: Application to Situations of Substrate Contamination and Background Subtraction." *Anal. Biochem. 184:*274-278.

Lee, J.Y, K.H. Jung, S.H. Choi, and H.S. Kim. 1995. "Combination of the *tod* and the *tol* Pathways in Redesigning a Metabolic Route of *Pseudomonas putida* for the Mineralization of a Benzene, Toluene, and *p*-Xylene Mixture." *Appl. Environ. Microbiol. 61*(6):2211-2217.

Lefever, M.R., and L.P. Wackett. 1994. "Oxidation of Low Molecular Weight Chloroalkanes by Cytochrome $P450_{CAM}$." *Biochem. and Biophysical Research Communications 201*(1):373-378.

Leson, G., and A.M. Winer. 1991. "Biofiltration: An Innovative Air Pollution Control Technology for VOC Emissions." *J. Air Waste Manage. Assoc. 41*(8):1045-1054.

Leson, G., and B.J. Smith. 1995. "Results from the PERF Field Study on Biofilters for Removal of Volatile Petroleum Hydrocarbons." *Proceedings of the 1995 Conference on Biofiltration*, University of Southern California, October 5-6, pp. 99-113.

Leson, G., D.S. Hodge, F. Tabatabai, and A.M. Winer. 1993. "Biofilter Demonstration Projects for the Control of Ethanol Emissions." Paper no. 93-WP-52C.04, 86th Annual Air & Waste Management Association Meeting & Exhibition, Denver, CO, June 13-18.

Li, S., and L.P. Wackett. 1993. "Reductive Dehalogenation by Cytochrome $P450_{CAM}$: Substrate Binding and Catalysis." *Biochemistry 32*:9355-9361.

Little, C.D., A.V. Palumbo, S.E. Herbes, M.E. Lidstrom, R.L. Tyndall, and P.J. Gilmer. 1988. "Trichloroethylene Biodegration by a Methane-Oxidizing Bacterium." *Appl. Environ. Microbiol. 54*(4):951-956.

Loy, J. 1995. "Biological Elimination of Odoriferous Pollutants and Solvents in Waste Gas with the Biotrickling Filter." Paper no. 95-MP9A.06, 88th Annual Air & Waste Management Association Meeting & Exhibition, San Antonio, TX, June 18-23.

Mackay, D., and W.Y.Shiu. 1981. "Critical Review Of Henry's Law Constants For Chemicals Of Environmental Interest." *J. Phys. Ref. Data 10*(4):1175-1199.

Malachowsky, K.J., T.J. Phelps, A.B. Teboli, D.E. Minnikin, and D.C. White. 1994. "Aerobic Mineralization of Trichloroethylene, Vinyl Chloride, and Aromatic Compounds by *Rhodococcus Species.*" *Appl. Environ. Microbiol. 60*(2):542-548.

McFarland, M.J., C.M. Vogel, and J.C. Spain. 1992. "Methanotrophic Cometabolism of Trichloroethylene (TCE) in a Two Stage Bioreactor System." *Wat. Res. 26*(2):259-265.

McGrath, M.S., and S.J. Ergas. 1995. "Hollow Fiber Membrane Bioreactor for Control of Volatile Organic Compound Emissions." Paper no. 95-TP9C.03, 88th Annual Air & Waste Management Association Meeting & Exhibition, San Antonio, TX, June 18-23.

Mu, D.Y., and K.M. Scow. 1994. "Effect of Trichloroethylene (TCE) and Toluene Concentrations on TCE and Toluene Biodegradation and the Population Density of TCE and Toluene Degraders in Soil." *Appl. Environ. Microbiol. 60*(7):2661-2665.

Nelson, M.J.K., S.O. Montgomery, W.R. Mahaffey, and P.H. Pritchard. 1987. "Biodegradation of Trichloroethylene and Involvement of an Aromatic Pathway." *Appl. Environ. Microbiol. 53*:949-954.

Nelson, M.J.K., S.O. Montgomery, and P.H. Pritchard. 1988. "Trichloroethylene Metabolism by Microorganisms that Degrade Aromatic Compounds." *Appl. Environ. Microbiol. 54*(2):604-606.

Nelson, M.J.K.., S.O. Montgomery, E.J. O'Neill, and P.H. Pritchard. 1986. "Aerobic Metabolism of Trichloroethylene by a Bacterial Isolate." *Appl. Environ. Microbiol. 58*(2):383-384.

Nishino, S.F., J.C. Spain, L.A. Belcher, and C.D. Litchfield. 1992. "Chlorobenzene Degradation by Bacteria Isolated from Contaminated Groundwater." *Appl. Environ. Microbiol. 58*(5):1719-1726.

Ockeloen, H.F., T.J. Overcamp, and C.P.L. Grady, Jr. 1992. "A Biological Fixed-Film Simulation Model for Removal of Volatile Organic Pollutants." Paper no. 92-116.05, 85th Annual Air & Waste Management Association Meeting & Exhibition, Kansas City, MO, June 21-26.

Oldenhuis, R. 1992. "Microbial Degradation of Chlorinated Compounds: Application of Specialize Bacteria in the Treatment of Contaminated Soil and Waste Water." PhD. Thesis. Rijksuniversiteit Groningen, Groningen, The Netherlands.

Oldenhuis, R., J.Y. Oedzes, J.J. van der Waarde, and D.B. Janssen. 1991. "Kinetics of Chlorinated Hydrocarbon Degradation by *Methylosinus trichosporium* OB3b and Toxicity of Trichloroethylene." *Appl. Environ. Microbiol. 57*:7-14.

Oldenhuis, R., R.L.J.M. Vink, D.B. Janssen, and B. Witholt. 1989. "Degradation of Chlorinated Aliphatic Hydrocarbons by *Methylosinus trichosporium* OB3b Expressing Soluble Methane Monooxygenase." *Appl. Environ. Microbiol. 55*(11):2819-2826.

Ottengraf, S.S.P. 1987. "Method for Biological Treatment of Waste Gases." U.S. patent no. 4,662,900.

Overcamp, T.J., B.F. Smets, R.E. Hammervold, and C.P.L. Grady, Jr. 1994. "A Bioregenerated Sorptive Slurry Scrubber." In *49th Purdue Industrial Waste Conference Proceedings*, Lewis Publishers, Chelsea, MI, pp. 419-424.

Overcamp, T.J., H.C. Chang, and C.P.L. Grady, Jr. 1993. "An Integrated Theory For Suspended Growth Bioscrubbers." *J. Air Waste Manage. Assoc. 43*(5):753-759.

Overcamp, T.L., H.F. Ockeloen, H.C. Chang, and C.P.L. Grady, Jr. 1992. "Design Criteria for Bioscrubbers: Fixed-Film Versus Suspended-growth Reactors." Presented at the 9th World Clean Air Conference and Exhibition, Montreal, Quebec, Canada, Aug. 30 - Sept. 4.

Pedersen, A.R., and E. Arvin. 1995. "Removal of Toluene in Waste Gases Using a Biological Trickling Filter." *Biodegradation 6*:109-118

Peters, D.A., G.T. Hickman, J.G. Stefanoff, and M.B.Garcia, Jr. 1993. "Laboratory Assessment of Biofiltration for Fuel-derived VOC Emissions Control." Paper no. 93-WA-52B.06, 86th Annual Air & Waste Management Association Meeting & Exhibition, Denver, CO, June 13-18.

Pettigrew, C.A., B.E. Haigler, and J.C. Spain. 1991. "Simultaneous Biodegradation of Chlorobenzene and Toluene by a *Pseudomonas* Strain." *Appl. Environ. Microbiol. 57*(1):157-162.

Phelps, T.J., J.J. Neidzielski, R.M. Schram, S.E. Herbes, and D.C. White. 1990. "Biodegradation of Trichloroethylene in Continuous-Recycle Expanded-Bed Bioreactors." *Appl. Environ. Microbiol.* *56*(6):1702-1709.

Phelps, T.J., K. Malachowsky, R.M. Schram, and D.C. White. 1991. "Aerobic Mineralization of Vinyl Chloride by a Bacterium of the Order *Actinomycetales*." *Appl. Environ. Microbiol.* *57*(4):1252-1254.

Radian/Envirogen. 1996. Final report for DoD contract F41624-95-C-8016, "2-Phase Extraction and Gas-phase Bioreactor Treatment of TCE in Soil and Groundwater." In press.

Rasche, M.E., M.R. Hyman, and D.J. Arp. 1991. "Factors Limiting Aliphatic Chlorocarbon Degradation by *Nitrosomonas europaea*: Cometabolic Inactivation of Ammonia Monooxygenase and Substrate Specificity." *Appl. Environ. Microbiol.* *57*(10):2986-2994.

Reij, M.W., K.D. de Gooijer, J.A.M. de Bont, and S. Hartmans. 1995a. "Membrane Bioreactor with a Porous Hydrophobic Membrane as a Gas-Liquid Contactor for Waste Gas Treatment." *Biotechnol. Bioeng.* *45*:107-115.

Reij. M.W., J. Kieboom, J.A.M. de Bont, and S. Hartmans. 1995b. "Continuous Degradation of Trichloroethylene by *Xanthobacter* sp. Strain Py2 During Growth on Propene." *Appl. Environ. Microbiol.* *61*(8):2936-2942.

Revah, S., A. Hinojosa, E. Marroquin, and V. Morales. 1994. "Biotechnological Process for the Treatment of Hydrogen Sulfide and Carbon Disulfide in a Waste Gas." Presented at the First North American Conference on Emerging Clean Air Technologies and Business Opportunities. Toronto, Canada, September.

Revah, S., M. Acosta, W. Hugler, R. Trinidad, C. Avila, I. Estrada, and A. Hinojosa. 1995. "Air Biodesulfurization from Viscose Plants: Carbon Disulfide Elimination." *Proceedings of the 1995 Conference on Biofiltration*, University of Southern California, October 5-6, pp. 81-187.

Schraa, G., M.L. Boone, M.S.M Jetten, A.R.W. van Neerven, P.J.Colberg, and A.J.B. Zehnder. 1986. "Degradation of 1,4-Dichlorobenzene by *Alcaligenes* sp. Strain A175." *Appl. Environ. Microbiol.* *52*:1374-1381.

Selifonov, S.A., M. Grifoll, R.W. Eaton, and P.J. Chapman. 1996. "Oxidation of Naphthenoaromatic and Methyl-Substituted Aromatic Compounds by Naphthalene 1,2-Dioxygenase." *Appl. Environ. Microbiol.* *62*(2):507-514.

Semprini, L., G.D. Hopkins, P.V. Roberts, D. Grbi -Gali , and P.L. McCarty. 1991. "A Field Evaluation of In-Situ Biodegradation of Chlorinated Ethenes: Part 3, Studies of Competitive Inhibition." *Groundwater* *29*(2):239-250.

Semprini, L., P.V. Roberts, G.D. Hopkins, and P.L. McCarty. 1990. "A Field Evaluation of In-Situ Biodegradation of Chlorinated Ethenes: Part 2, Results of Biostimulation and Biotransformation Experiments." *Groundwater* *28*(5):715-727.

Shareefdeen, Z., B.C. Baltzis, Y-S. Oh, and R. Bartha. 1993. "Biofiltration Of Methanol Vapor." *Biotechnol. Bioeng. 41*:512-524.

Shields, M.S., and M.J. Reagin. 1992. "Selection of a Pseudomonas cepacia Strain Constitutive for the Degradation of Trichloroethylene." *Appl. Environ. Microbiol. 58*(12):3977-3983.

Shields, M.S., M.J. Reagin, R.R. Gerger, C. Somerville, R. Schaubhut, R. Campbell, and J. Hu-Primmer. 1993. "Constitutive Degradation of Trichloroethylene by an Altered Bacterium in a Gas Phase Bioreactor." Presented at the 1993 Battelle In-situ and On-site Bioreclamation Conference in San Diego, CA.

Shields, M.S., S.O. Montgomery, P.J. Chapman, S.M. Cruskey, and P.H. Pritchard. 1989. "Novel Pathway of Toluene Catabolism in the Trichloroethylene-degrading Bacterium G4." *Appl. Environ. Microbiol. 55*:1624-1629.

Smits, M.C.J., S.P.P. Ottengraf, and J.C. van den Heuvel. 1995. "Effect of the Superficial Liquid Velocity on the Nitrification of Ammonia Polluted Air in a Bio-Trickling Filter." *Proceedings of the 1995 Conference on Biofiltration*, University of Southern California, October 5-6, pp. 199-206.

Sorial, G.A., F.L. Smith, A Pandit, M.T. Suidan, P. Biswas, and R.C. Brenner. 1995. "Performance of Trickle Bed Biofilters Under High Toluene Loading." Paper no. 95-TA9B.04, 88th Annual Air & Waste Management Association Meeting & Exhibition, San Antonio, TX, June 18-23.

Sorial, G.A., F.L. Smith, M.T. Suidan, P. Biswas, and R.C. Brenner. 1994. "Evaluation of the Performance of Trickle Bed Biofilters - Impact of Periodic Removal of Accumulated Biomass." Paper no. 94-RA115A.05, 87th Annual Air & Waste Management Association Meeting & Exhibition, Cincinnati, OH, June 19-24.

Spain, J.C., and S.F. Nishino. 1987. "Degradation of 1,4-Dichlorobenzene by a *Pseudomonas* sp." *Appl. Environ. Microbiol. 53*(5):1010-1019.

Speitel, G.E. Jr., and J.M. Leonard. 1992. "A Sequencing Biofilm Reactor for the Treatment of Chlorinated Solvents Using Methanotrophs." *Water Environ. Res. 64*(5):712.

Speitel, G.E., and D.S. McClay. 1993. "Biofilm Reactors for Treatment of Gas Streams Containing Chlorinated Solvents." *ASCE J. Envir. Engrg. 119*(4):658-678.

Spiess, E., C. Sommer, and H. Grisch. 1995. "Degradation of 1,4-Dichlorobenzene by *Xanthobacter flavus* 14p1." *Appl. Environ. Microbiol. 61*(11):3884-3888.

Strand, S.E., M.D. Bjelland, and H.D. Stensel. 1990. "Kinetics of Chlorinated Hydrocarbon Degradation by Suspended Cultures of Methane-Oxidizing Bacteria." *Res. J. WPCF. 62*(2):124-129.

Strotmann, U.J. M. Pentegna, and D.B. Janssen. 1990. "Degradation of 2-Chloroethanol by Wild Type and Mutants of *Pseudomonas putida* US2." *Arch. Microbiol. 154*:294-300.

Sutton, P.M., and P.N. Mishra. 1994. "Activated Carbon Based Biological Fluidized Beds for Contaminated Water and Wastewater Treatment: A State-of-the-Art Review." *Wat. Sci. Tech.* *29*(10-*11*:309-317.

Togna, A.P., and B.R. Folsom. 1992. "A Comparative Study of Biofilter and Biotrickling Filter Performance for Isopentane Removal." Paper no. 92-116.04, 85th Annual Air & Waste Management Association Meeting & Exhibition, Kansas City, MO, June 21-26.

Togna, A.P., and M. Singh. 1994. "A Comparative Study of Biofilter and Biotrickling Filter Performance for Isopentane Removal." Paper no. 94-RP115B.04, 87th Annual Air & Waste Management Association Meeting & Exhibition, Cincinnati, OH, June 19-24.

Togna, A.P., G.J. Skladany, J.M. Caratura, and S. Frisch. 1993. "Field-Pilot Results of Styrene Biodegradation Using Biofiltration: A Case Study." In *48th Purdue Industrial Waste Conference Proceedings*, Lewis Publishers, Chelsea, MI, pp. 517-527.

Togna, A.P., G.J. Skladany, W.J. Fucich, W.J. Guarini, M. Singh, and T.S. Webster. 1995. "Removal of Isopentane and Isobutane Vapors from an Industrial Process Waste Stream Using a Field-pilot Biotrickling Filter." Paper no. 95-TA9B.02, 88th Annual Air & Waste Management Association Meeting & Exhibition, San Antonio, TX, June 18-23.

Togna, A.P., G.S. Skladany, and J.M. Caratura. 1994. "Treatment of BTEX and Petroleum Hydrocarbon Vapors Using a Field-Pilot Biofilter." In *49th Purdue Industrial Waste Conference Proceedings*, Lewis Publishers, Chelsea, MI, pp. 437-448.

Torres-Cardona, M.D., S. Revah-Moiseev, A. Hinjosa-Martinez, F.J. Paez-Moreno, and V.M. Morales-Baca. 1993. "Biological Process or the Elimination of Sulfur Compounds Present in Gas Mixtures." U.S. patent no. 5,236,677.

Tsien, H.C., G.A. Brusseau, R.S. Hanson, and L.P. Wackett. 1989. "Biodegradation of Trichloroethylene by *Methylosinus trichosporium* OB3b." *Appl. Environ. Microbiol.* *55*(12):3155-3161.

U.S. Department of Energy (DoE). 1994. *Field Demonstration of Vapor Phase TCE Bioreactor.* DOE/OR/21400-T492, prepared for the U.S. Department of Energy.

van den Wijngaard, A.J., K.W.H.J. Van Der Kamp, J. Van Der Ploeg, F. Pries, B. Kazemier, and D.B. Janssen. 1992. "Degradation of 1,2-Dichloroethane by *Ancylobacter aquaticus* and Other Facultative Methylotrophs." *Appl. Environ. Microbiol. 58*(3):976-983.

van den Wijngaard, A.J., R.D. Wind, and D.B. Janssen. 1993. "Kinetics of Bacterial Growth on Chlorinated Aliphatic Compounds." *Appl. Environ. Microbiol. 59*(7):2041-2048.

van der Ploeg, J., M.P. Smidt, A.S. Landa, and K.B. Janssen. 1994. "Identification of Chlorocataldehyde Dehydrogenase Involved in 1,2-Dichloroethane Degradation." *Appl. Environ. Microbiol. 60*(5):1599-1605.

van Groenestijn, J.W., and P.G.M. Hesselink. 1993. "Biotechniques for Air Pollution Control." *Biodegradation 4*:283-301.

van Lith, C., S.L. David, and R. Marsh. 1990. "Design Criteria for Biofilters." *Trans. IChemE. 68*(Part B):127-132.

van Lith, C.P.M., S.P.P. Ottengraf, and R.M.M. Diks. 1994. *Proceedings of the International Symposium on Biological Waste Gas Cleaning.* Heidelberg, Germany, March 9-11. VDI Berichte No. 1104:169-180.

van Lith, C.P.M., S.P.P. Ottengraf, B. Osinga, and R.M.M. Diks. 1993. "The Biological Purification of a Multi-Component Waste-Gas Discharged in Artificial Glass Production." Paper no. 93-WA-52B.07, 86th Annual Air & Waste Management Association Meeting & Exhibition, Denver, CO, June 13-18.

Vannelli, T., M. Logan, D.M. Arciero, and A.B. Hooper. 1990. "Degradation of Halogenated Aliphatic Compounds by the Ammonia-Oxidizing Bacterium *Nitrosomonas europaea*." *Appl. Environ. Microbiol. 56*(4):1169-1171.

VDI Richtlinie 3477. 1991. *Biological Waste Gas/Waste Air Purification, Biofilters.* Dusseldorf, VDI Handbuch Reinhaltung der Luft, Band 6.

VDI Richtlinie 3478. 1985. *Biological Waste Air Purification, Bioscrubbers.* Dusseldorf, VDI Handbuch Reinhaltung der Luft, Band 6.

Vembu, K., and C.S. Walker. 1995. "Biofiltration Holds VOC Odors at Bay." *Environmental Protection,* February, p. 27.

Vogel, T.M., C.S. Criddle, and P.L. McCarty. 1987. "Transformations of Halogenated Aliphatic Compounds." *Environ. Sci. Technol. 21*(8):722-736.

Wackett, L.P,. G.A. Brusseau, S.R. Householder, and R.S. Hanson. 1989. "Survey of Microbial Oxygenases: Trichloroethylene Degradation by Propane-Oxidizing Bacteria." *Appl. Environ. Microbiol. 55*(11):2960-2964.

Wackett, L.P., and D.T. Gibson. 1988. "Degradation of Trichloroethylene by Toluene Dioxygenase in Whole-Cell Studies with *Pseudomonas putida* F1." *Appl. Environ. Microbiol. 54*(7):1703-1708.

Wackett, L.P., and S.R. Householder. 1989. "Toxicity of Trichloroethylene to *Pseudomonas putida* F1 is Mediated by Toluene Dioxygenase." *Appl. Environ. Microbiol. 55*(10):2723-2725.

Wackett, L.P., M.J. Sadowsky, L.M. Newman, H.G. Hur, and S. Li. 1994. "Metabolism of Polyhalogenated Compounds by a Genetically Engineered Bacterium." *Nature 368*:627-629.

Wark, K and C.F. Warner. 1981. *Air Pollution: Its Origin and Control*, 2nd Ed. Harper and Row, New York, pp. 405-423.

Weber, F.J., and S. Hartmans. 1994. "Toluene degradation in a trickle bed reactor - Prevention of clogging." *Proceedings of the International Symposium on Biological Waste Gas Cleaning*, Heidelberg, Germany, March 9-11. VDI Berichte No. 1104:161-168.

Webster, T.S., J.S. Devinny, E.M. Torres, S.S. Basrai, and V. Kogan. 1995. "Study of Biofiltration for Control of VOC and Toxics Emissions from Wastewater Treatment Plants: Phase II Bench-scale Experiments." Paper no. 95-TA9B.06, 88th Annual Air & Waste Management Association Meeting & Exhibition, San Antonio, TX, June 18-23.

Wiedemeier, T., J.T. Wilson, D.H. Kampbell, R.N. Miller, and J.E. Hansen. 1995. "Technical Protocol for Implementing Intrinsic Remediation with Long-Term Monitoring for Natural Attenuation of Fuel Contamination in Groundwater." Prepared for the U.S. Air Force Center for Environmental Excellence (AFCEE), Technology Transfer Division, Brooks AFB, San Antonio, Texas.

Wilson, J.T., and B.H. Wilson. 1985. "Biotransformation of Trichloroethylene in Soil." *Appl. Environ. Microbiol.* 49:242-243.

Yang, Y., and D. Alibeckoff. 1995. "Biofiltration for Control of Carbon Disulfide and Hydrogen Sulfide Vapors." *Proceedings of the 1995 Conference on Biofiltration*, University of Southern California, October 5-6, pp. 165-179.

Yang, Y., and E.R. Allen. 1994a. "Biofiltration Control of Hydrogen Sulfide 1. Design and Operational Parameters." *J. Air Waste Manage. Assoc. 44*(7):863-868.

Yang, Y., and E.R. Allen. 1994b. "Biofiltration Control of Hydrogen Sulfide 1. Kinetics, Biofilter Performance, and Maintenance." *J. Air Waste Manage. Assoc. 44*(11):1315-1321.

Yang, Y., A.P. Togna, and G. Skladany. 1994. "Treatment of CS_2 and H_2S Vapors by Biofiltration." In *49th Purdue Industrial Waste Conference Proceedings*, Lewis Publishers, Chelsea, WI, pp. 449-456.

Yang, Y., A.P. Togna, and J.R. Blunk. 1993a. "Treatment of Carbon Disulfide by Biofiltration." Presented at the 1993 Superfund XIV Conference & Exhibition, Washington, D.C., Nov. 30-Dec. 2.

Yang, Y., J.M. Caratura, and D. Empfield. 1993b. "Use of Biofiltration to Control Hydrogen Sulfide Emissions." Presented at the 16th International Symposium on Gas, Oil, and Environmental Biotechnology, Colorado Springs, CO, Nov. 29 - Dec. 1.

Yavorsky, J.. 1993. "Biofiltration for Control of Gas Streams Containing Low Concentrations of Volatile Organic Compounds." Paper no. 93-RP-139.02, 86th Annual Air & Waste Management Association Meeting & Exhibition, Denver, CO, June 13-18.

Ye, L., N.N. Khandan, and F. G. Edwards. 1994. "Biological Treatment of Airstreams Contaminated with Organic Vapors." *Wat. Sci. Tech. 30*(7):71-74.

Zahodiakin, P. 1990. "Puzzling Out the New Clean Air Act." *Chem. Engr. 97*(12):24-27.

Zurlinden, R.A., J.C. Lucas and C. Carmel. 1994. "Control of Gasoline-Derived Volatile Organic Compounds by Biofiltration." Paper no. 94-RA115A.06, 87th Annual Air & Waste Management Association Meeting & Exhibition, Cincinnati, OH, June 19-24.

Zylstra, G.J., L.P. Wackett, and D.T. Gibson. 1989. "Trichloroethylene Degradation by *Escherishia coli* Containing the *Pseudomonas putida* F1 Toluene Dioxygenase Genes." *Appl. Environ. Microbiol.* *55*(12):3162-3166.

APPENDIX A
LABORATORY WORK PLAN

This work plan describes the approach for the laboratory phase of the McClellan AFB Biological Treatment of SVE Off-Gas demonstration. The work plan describes the system configuration and operation, method of introducing the vapor-phase contaminants to the reactors, and the analytical protocol, including sampling frequency and methods.

A.1 Development of a Contaminated Vapor Phase to Simulate the SVE Off-Gas Contaminants

Six VOCs were selected from the contaminants detected in the SVE off-gas for the laboratory demonstration (Table A.1). They include PCE; TCE; 1,1,1-TCA; 1,2-DCB; toluene; and acetone. The gas-phase concentrations of the contaminants will be similar to their average concentrations detected in the SVE off-gas stream and shown in Table A.1. The contaminants will be injected into a gas mixing chamber, upstream of the reactors, using a syringe pump. Makeup gas will be provided by a pressurized air cylinder at the desired gas flowrate.

TABLE A.1. CONTAMINANTS SELECTED FOR THE LABORATORY DEMONSTRATION

	Off-Gas Conc.		
Compound	Max. (ppmv)	Avg. (ppmv)	H2O Concentration (mg/L)
PCE	97.4	63.0	0.403
TCE	111.3	74.4	1.07
1,1,1-TCA	241.4	152.9	4.15
1,2-DCB	120.6	35.5	2.82
Toluene	76.8	45.5	0.657
Acetone	92.0	63.8	180
Chlorinated Sum	571	326	8.44
Nonchlorinated Sum	169	110	180

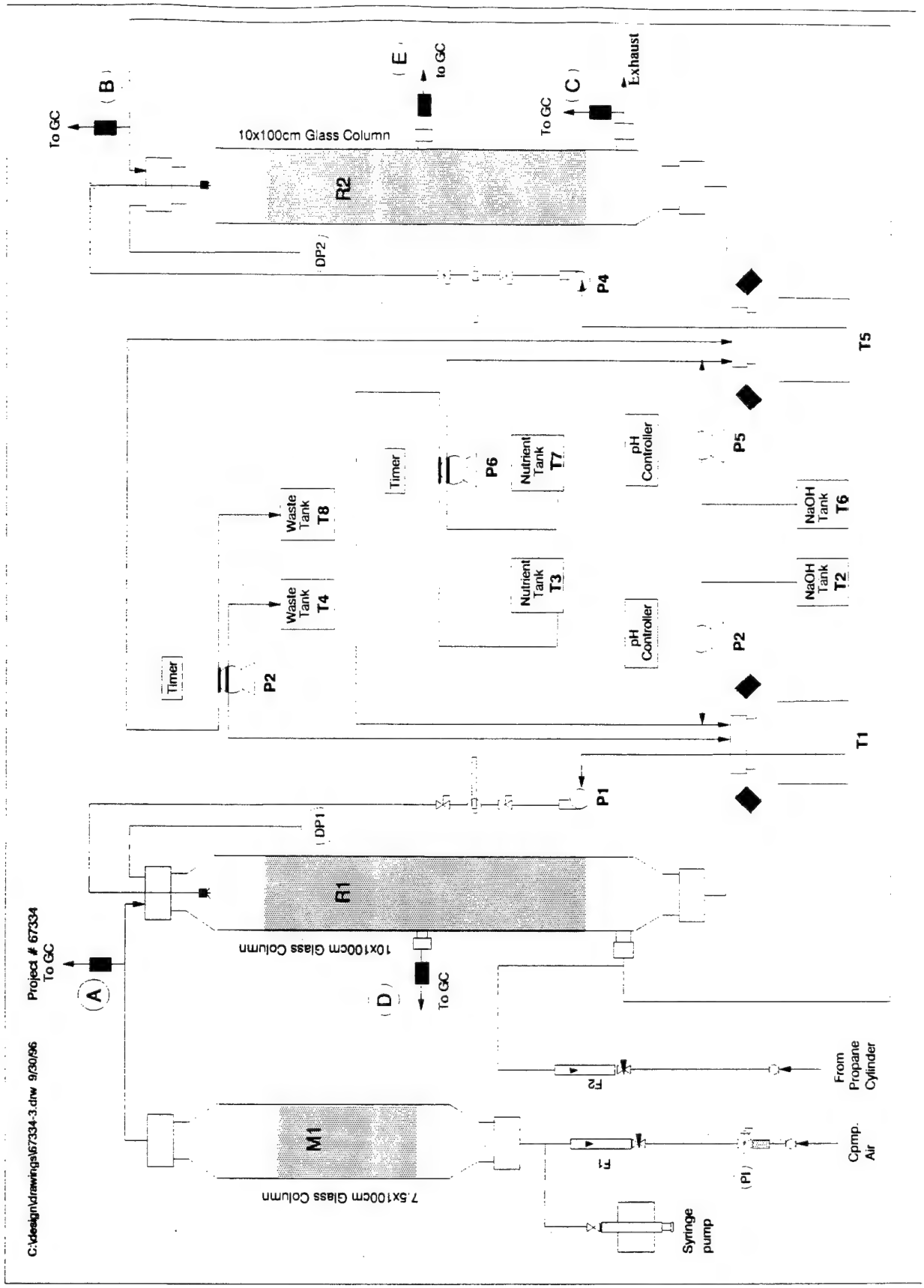

Figure A.1. Schematic Drawing of the Biotrickling Filter System 1 Propane Cosubstrate

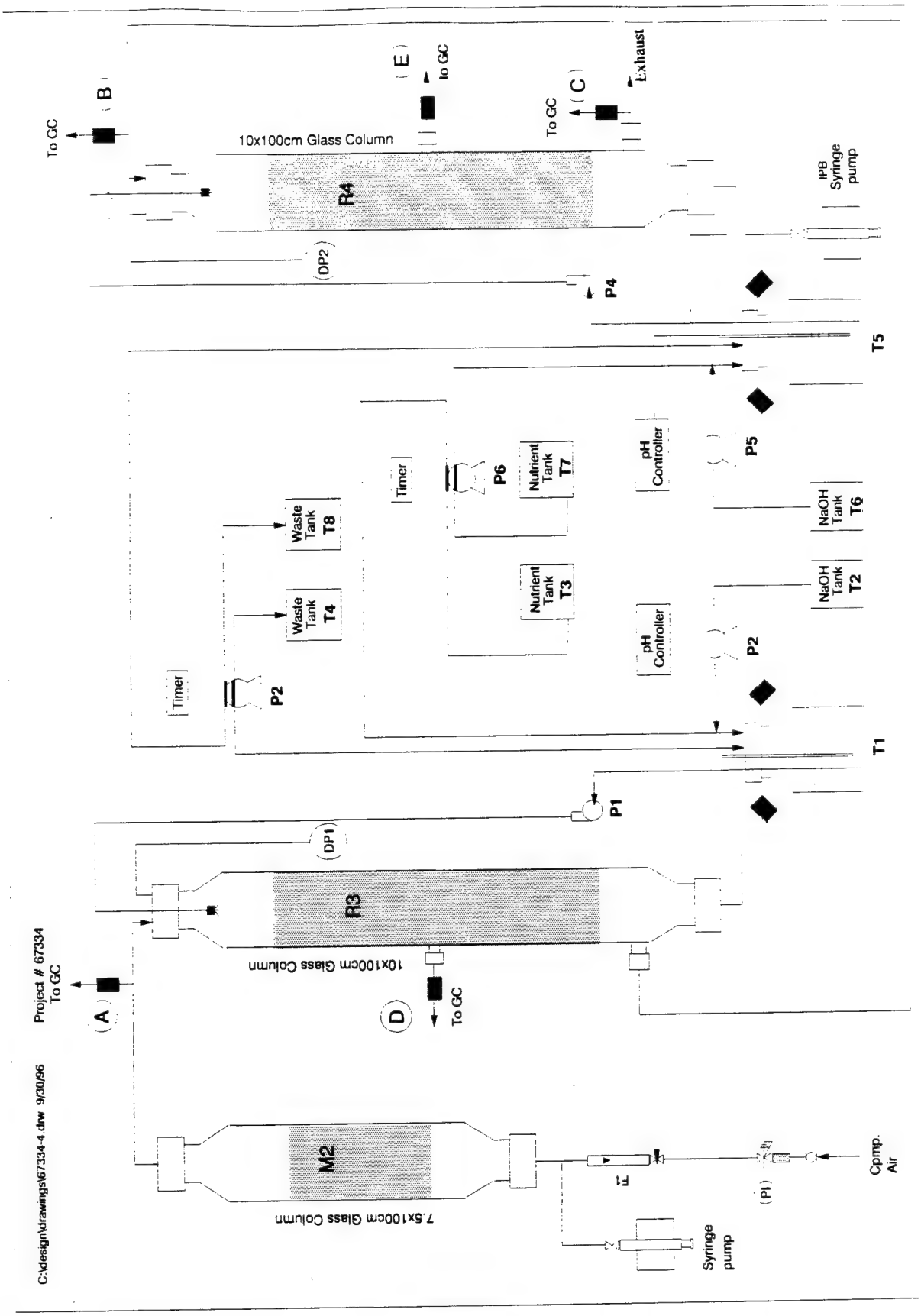

Figure A.2. Schematic drawing of the Biotrickling Filter System 2 Isopropylbenzene Cosubstrate

The two-stage laboratory-scale reactor system is shown schematically in Figure A.1 and Figure A.2. Cosubstrates (propane or IPB) will be fed to the second stage of each system. Propane will be added through a gas flowmeter and will be monitored in the gas phase at the beginning and end of the second reactor stage to monitor the mass of propane consumed in the reactor. IPB will be added in the aqueous phase with the nutrients and will be metered into the reactor liquid phase using a peristaltic pump.

The reactors will consist of 10 cm by 100 cm glass cylinders operated sequentially. The contaminant gas mixture will be fed to the reactors from an air-sparged contaminated aqueous solution. After the reactors are set up, they will be operated abiotically, to shake down the system for possible leaks, to determine whether there are abiotic losses in the system in the absence of microbial growth, and to assess the recovery of the contaminants in the system. The reactors will be operated without biomass for approximately 4 days.

The second stage of the filter system will be inoculated with propane-oxidizing bacteria and IPB-oxidizing bacteria that will be obtained from Envirogen's laboratories, or other laboratories, to accelerate the growth and acclimation period during startup. Conversely, the first stage reactors will be inoculated with a general microbial consortium (Phenobac®, Polybac, Bethlehem, PA).

Initially, the vapor flowrate through each system will be set to establish a vapor contact time of approximately 4 to 5 minutes in each stage. The system will be optimized in the laboratory for lower vapor contact times.

A.2 Reactor Sampling

Influent and effluent vapor concentrations will be monitored using an on-line gas chromatograph (GC). Sampling ports will be located at (1) the influent to each system; (2) the effluent from the first stage of each system; and (3) the effluent from the second stage of each system and (4 and 5) the center of the two reactor stages. Sampling ports will be configured to permit periodic grab sampling. The online GC will sample the reactor influent and effluent streams twice daily to provide gas-phase contaminant concentrations. The intermediate port samples will provide information on the degradation characteristics within the columns.

Once steady state is established, liquid samples will be taken in triplicate from each reactor stage on a weekly basis throughout the duration of the laboratory demonstration. The liquid samples will be

analyzed for aqueous-phase organic contaminants using EPA Method 8260 and a GC/MS equipped with a purge-and-trap. Total organic carbon (TOC) will be measured using EPA Method 415.1. The purpose of the liquid sampling will be to establish careful mass balances of the contaminants and cosubstrates in the reactors during the study. Propane concentrations will be monitored in the vapor phase only, while IBP concentrations will be monitored in the aqueous phase, which will require more frequent samples to maintain target effluent IPB concentrations. Twice a week, liquid samples will be analyzed for chloride concentrations using an ion-selective probe. The pH will be monitored continuously using a dedicated probe in the reactors.

A.3 System Performance Under Stressed Conditions

Steady state will be established when effluent gas contaminant and cosubstrate concentrations remain constant over time, (i.e. ±20% for 4 days). Once steady state operating conditions are established, the system will be analyzed to evaluate its ability to handle shock loads. Shock loads will be imposed by rapidly (i.e., instantaneously) increasing the simulated off-gas flow rates or off-gas contaminant concentrations. The shock loads will be designed to mimic the full-scale system by increasing the reactor mass flowrates by the ratio of maximum to average off-gas contaminant concentrations, shown in Table A.1. During the stress tests, the online monitoring frequency of the reactors will be increased. Cosubstrate feed concentrations will be increased to maintain a constant cosubstrate/CAC feed ratio. If the biological reactors are able to handle the shock loads without adverse effects, the cosubstrate concentrations will be reduced while maintaining high gas contaminant concentrations to evaluate reactor performance under those conditions. Reactors will be sampled twice daily during the stress tests.

A.4 System Optimization

For the first two months of operation, no attempt will be made to optimize the system, although the cosubstrate/CAC ratios may be adjusted, if needed, to increase performance, and ethanol may be added to the 1st stage of each system to increase biofilm growth. In addition, a small amount of PAC may be added to the 2nd stage of each system to better regulate the supply of cosubstrate to the microorganisms. After the first two months, once steady-state has been achieved, the system will be optimized by increasing the vapor throughput until the predetermined DREs are no longer achieved; this will be the maximum vapor throughput. As the vapor flowrate is increased, the cosubstrate addition rate will be adjusted to maintain a constant cosubstrate/CAC ratio. Cosubstrate feeding also will be

optimized by decreasing the cosubstrate/CAC ratio while maintaining the minimum DREs to establish the minimum possible cosubstrate/CAC ratio. The optimization period is expected to last for approximately 1.5 months. During that period, vapor and liquid sampling frequencies will be the same as described for the steady state operating period.

A.5 Quality Assurance

Envirogen's laboratory is certified by the New Jersey Department of Environmental Protection (NJDEP) for Method 8260 and 415.1 analyses. For these analyses, all laboratory quality assurance/quality control (QA/QC) practices will be in accordance with NJDEP's Laboratory Certification Program. For the online vapor analyses, check standards with known concentrations will be analyzed weekly. The GC system will be calibrated if the measurements fall outside ±20% of the known check standard concentrations. In addition, influent and effluent vapor samples will be collected at two time points using evacuated canisters for Method T0-14 analyses by an independent outside laboratory, selected by Battelle. Results will be compared to the online analyses. As a QA/QC check of the chloride measurements using the a chloride probe, the measurements from the probe will be compared with ion chromatography measurements performed by Envirogen's laboratory (Method 300), at four time points.

APPENDIX B

CALCULATIONS

$$\text{Mass Flow Rate}\left[\text{lb / hr}\right] = C \cdot \left(\frac{\text{MW}}{22.4 \cdot 10^{-3} \ \frac{m^3}{mol}} \right) \cdot \left(\frac{273}{\text{T}} \right) \cdot \left(\frac{\text{P}}{1 \text{ atm}} \right) \cdot \bar{Q} \cdot \text{B}$$

C = concentration [ppmv]

MW = molecular weight [g/mol]

T = ambient temperature [K] (298 degrees K)

P = ambient pressure [atm] (1 atm)

$\bar{Q}$ = average flowrate [ft^3/min]

B = conversion factor

$$\text{B} = \left(\frac{1 \text{ lb}}{453.6 \text{ g}} \right) \cdot \left(\frac{0.02832 \text{ m}^3}{\text{ft}^3} \right) \cdot \left(\frac{60 \text{ min}}{1 \text{ hr}} \right) \cdot \frac{\left(\frac{1 \text{ m}^3}{10^6 \text{ m}^3} \right)}{1 \text{ ppmv}}$$

22.4E-3 m^3/ mol - 1 mole of an ideal gas at 0°C and 1 atm pressure occupies a volume of 22.4E-3m^3

$$\text{Concentration in Water}\left[\text{mg / L}\right] = \frac{\text{C} \cdot \left(\frac{10^{-6} \text{atm}}{1 \text{ ppmv}} \right) \cdot \text{MW}}{\text{H} \cdot 1000 \ \frac{\text{L}}{\text{m}^3}} \cdot 1000 \text{ mg/g}$$

C - concentration [ppmv]

MW = molecular weight [g/mol]

H = Henry's Law Constant [atm • m^3/ mol]

Assumptions:

- Minimum, maximum, and average concentration calculations did not include samples with concentrations below the detection limits.

C:mswd\litrevR2.doc\VSM:cam